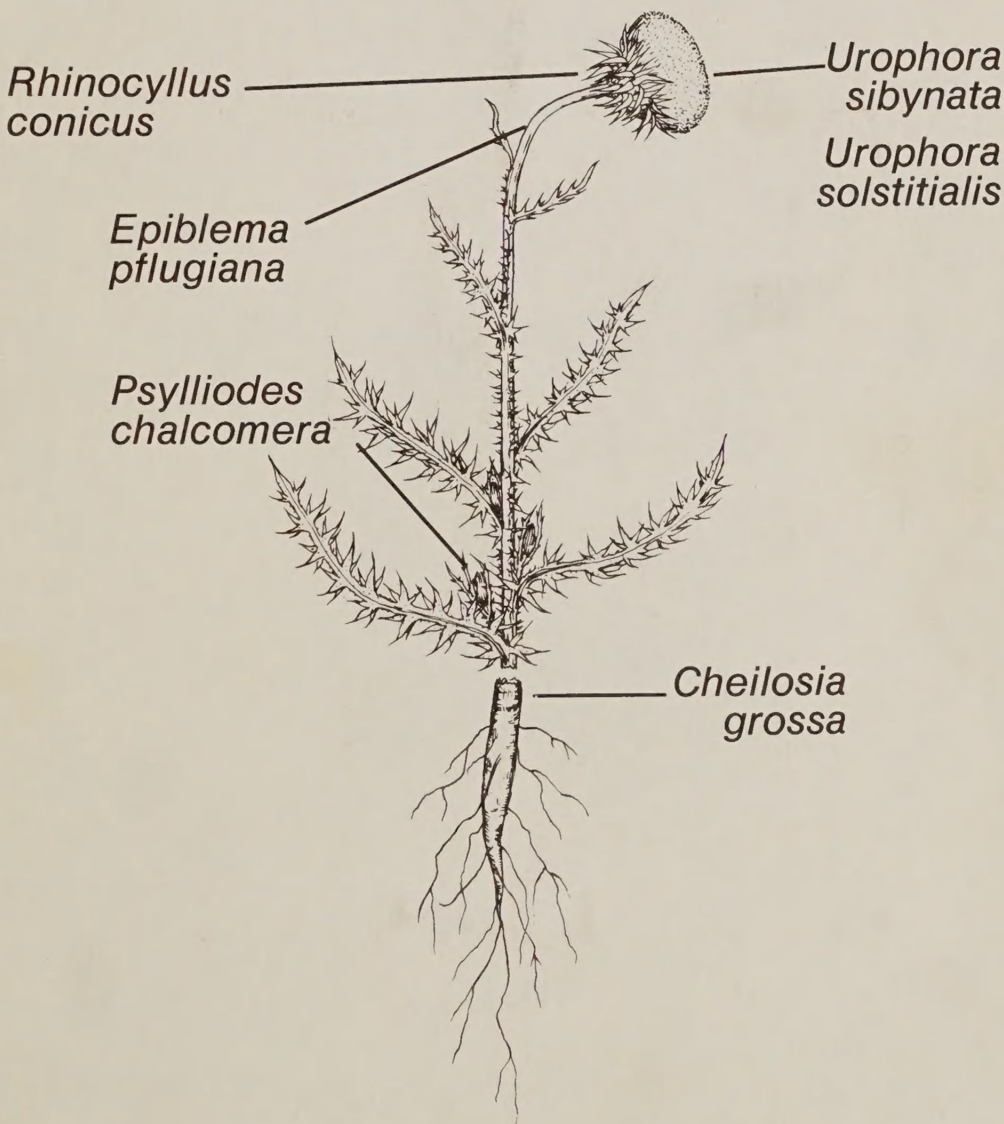
BIOLOGICAL CONTROL OF THISTLES in the Genus *Carduus* in the United States



A Progress Report

Science and Education Administration U.S. Department of Agriculture

Biological Control of Thistles

in the Genus *Carduus* in the United States

A Progress Report

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COVER: Several insects that attack the bolting stem or flower head of *Carduus macrocephalus* are potential agents for controlling this thistle. See pp. 11-17.

FOREWORD

Using insects to successfully control several thistles in the Eurasian genus *Carduus* appears to be in the offing in the United States. The exact number of large-flowered species involved under the name of musk thistle, *C. nutans* L., is presently unknown but is under study by Dr. M. K. McCarty, whose paper appears herein. However, the small-flowered species, plumeless thistle, *C. acanthoides* L., and welted thistle, *C. crispus* L., are distinct and are considered separately by various workers (Dunn 1976, Harris and Zwölfer 1971). There are two species in the slender-flowered group: Italian thistle, *C. pycnocephalus* L., and slenderflower thistle, *C. tenuiflorus* Curt., which because of their similarity are sometimes treated together; for example, as Italian thistle in California (Dunn 1976) or as slender thistle in Tasmania (Bendall 1975).

Following introduction into the United States, all of these species have either become pests or are rated as potential pests (Dunn 1976). For example, the musk thistle complex is now found in 40 of the adjacent 48 states, with 12% of the counties in those states rating their musk thistle infestations as economic. Plumeless thistle is found in 19 states, Italian thistle is widespread in California, and welted thistle is found in 51 counties of Virginia (Dunn 1976).

When a weed becomes widespread, as have several species of *Carduus* in Canada and the United States, effective, low-cost, and long-lasting controls are needed and all alternative methods are considered, including biological control. Therefore, a biological control program for selected weedy thistles, including *Carduus* spp., began in 1961 with a systematic survey of central Europe for phytophagous insects feeding on all species of thistles (Zwölfer 1965). These investigations were conducted through 1964 by the European station, Commonwealth Institute of Biological Control, and were sponsored by the Entomology Research Institute, Belleville, Ontario, Canada. Also, in the early 1960's, insect enemies of Italian thistle were being sought in the Mediterranean area for the United States (Holloway 1964). Subsequent comparison of surveys of areas where the thistles are considered to be indigenous vs. those where they are introduced indicated a discrepancy in the number of natural enemies between areas. In the case of Italian thistle, Goeden (1974) found about 33% of the insects feeding on it in southern Europe were endophagous in the immature stages (a criterion considered as indicating greater specialization and adaptation to the host plant), while in southern California about 12.5% were endophagous. As for musk thistle, Zwölfer (1965) listed about 43% of the European insects as endophagous, while in South Dakota there were only 3% so classified (Morihara and Balsbaugh 1976). Surveys conducted in Pakistan showed that 35% of the insect species were endophagous in the immature stages (Baloch et al. 1971). However, the Pakistan fauna lacked the species considered as most promising for biological control (Goeden 1974, Harris and Zwölfer 1971, Zwölfer et al. 1971).

As a result of these surveys and subsequent life history and host-plant specificity studies, the weevil *Rhinocyllus conicus* (Froelich) was introduced

and released in 1969 on musk and plumeless thistles in the United States in Montana (Rees 1977) and Virginia (Surles et al. 1974). Previously it had been released in 1968 in Canada (Harris and Zwölfer 1971) and subsequently in New Zealand in 1973 (Jessep 1975). A strain of this weevil, obtained from Italian thistle in southern Italy, was released in 1973 on Italian thistle in southern California (Goeden 1974). *R. conicus* established in Montana and Virginia (Andres and Davis 1973) and since that time has had an impact on the density of musk thistle both in Montana (Rees 1977) and in Virginia (Kok and Surles 1975). Because of these successes, approximately 90,000 *R. conicus* adults have since been released in about 20 more states (N. E. Rees, personal communication).

Realizing that additional insect enemies of these thistles probably would be required before adequate control over widespread areas could be achieved, several insects found during the surveys have been studied further (Dunn and Rizza 1976). Those selected include two weevils, *Ceuthorrhynchidius horridus* (Panzer) and *Ceutorhynchus trimaculatus* (F.), the larvae of which feed in the rootcrowns; a flea beetle, *Psylliodes chalcomera* (Illiger), the larvae of which feed in flower and leaf buds; and a syrphid fly, *Cheilosia* sp., the larvae of which feed in the stems and rootcrowns. All of these insects have been under study for several years, two of them, *P. chalcomera* (Dunn and Rizza 1976) and *C. horridus* (Kok 1975), extensively so.

The progress made through 1977 in the biological control of the weedy thistles in the genus *Carduus* was documented at a symposium held on November 28, 1977, at the National Meeting of the Entomological Society of America in Washington, D.C. Six of the papers presented herein were read at that time, while the seventh, by Dr. R. D. Goeden, who was unable to attend, was graciously prepared so that the biological control of the slender-flowered *Carduus* spp. in California could be included. An attempt has been made to present these papers in a logical sequence, beginning with a history of the project. This is followed by a discussion of the species included in the genus *Carduus*, particularly those in the large-flowered musk thistle complex. The remaining papers are concerned with the present state of progress, first in Europe, and then domestically in the northeastern states, Virginia, Montana, and California.

At present the prognosis appears to be favorable for success in controlling another weed by the use of insects. It is hoped that the biological control of the weedy species of *Carduus* will equal in degree that previously obtained for several weeds in the United States which have been suppressed to the point that their control has been rated as complete, substantial, or effective (Andres and Davis 1973). These more successful examples of biological weed control with insects include (1) St. Johnswort, *Hypericum perforatum* L., in the western United States, (2) alligatorweed, *Alternanthera philoxeroides* (Mart.) Griseb., in the southeastern United States, (3) tansy ragwort, *Senecio jacobaea* L., in northwestern California and western Oregon, (4) pricklypear cactus, *Opuntia* spp., in Hawaii and insular southern California, (5) puncture vine, *Tribulus terrestris* L., in Hawaii, (6) lantana, *Lantana camara* L., in Hawaii, and (7) pamakani, *Eupatorium adenophorum* Spr., in Hawaii (Andres and Davis 1973, Andres and Goeden 1971).

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HISTORY OF THE BIOLOGICAL CONTROL OF MUSK THISTLE IN NORTH AMERICA AND STUDIES WITH THE FLEA BEETLE PSYLLIODES CHALCOMERA

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The direct origins of the program for biological control of musk thistle are obscure. While tracing the history of this program, it became evident that the joint Canadian-American effort we know today started as separate, almost parallel American and Canadian projects, neither directed toward control of musk thistle. This paper will cover the origins of both projects and how they developed into one.

The oldest document I could find relating to biological control of Carduus thistle in the United States was a letter, dated October 24, 1956, to T. W. Gardner of the Insect Identification and Parasite Introduction Section, ARS, from James K. Holloway, head of the biological control of weeds studies, at Albany, California, and the USDA-ARS pioneer worker in this field. The letter concerned the abundance of Italian thistle in California, and pointed out that the name covered two closely related species, Carduus pycnocephalus L. and C. tenuiflorus Curt. The closing paragraph of the letter reads as follows: "Both species are annuals and in California have a relatively short growing season. Both of these characteristics would necessitate finding insects well synchronized with the plant growth phases because, to gain any measure of control, it is very likely the plant will have to be destroyed before it comes into flower. If this cannot be accomplished possibly a seed insect would be helpful."

That is where the biological control of Carduus thistles in the United States stood 21 years ago.

However, the program took on life in 1959, when the U.S. Department of Agriculture sent Dr. Lloyd Andres to Rome, Italy, to establish an overseas station for screening candidate insects for biological control of weeds. He had in his pocket a list of 19 weedy plants for which he was supposed to find natural enemies and clear their introduction into the United States. That list included yellow starthistle (Centaurea solstitialis L.), spotted knapweed (Centaurea maculosa Lam.), Canada thistle (Cirsium arvense (L) Scop.), and Italian thistle (Carduus pycnocephalus L.), this being the closest thing to musk thistle (Carduus nutans L.) on the list.

In 1960, in his broad-ranging surveys, Dr. Andres collected two weevils, Ceutorhynchus trimaculatus (F.) and Ceuthorrhynchidius horridus (Panz.); several species of Lixus; a tortoise beetle (Cassida sp.); and a phytophagous fly, probably a Cheilosia sp. (Syrphidae). In 1961, he added the weevil genus Larinus. All these insects were associated with Italian thistle, but no

particular attention was paid to musk thistle, and during 1962, it wasn't mentioned in the quarterly reports of the Rome laboratory.

In 1963, Dr. Kenneth Frick went to the USDA laboratory in Rome, and among his projects was to continue the survey of the insects attacking Italian thistle (Carduus pycnocephalus L.) and slenderflower thistle (Carduus tenuiflorus Curt.). He enlarged the project to include musk thistle (Carduus nutans L.) and milk thistle (Silybum marianum Gaertn.). While making this survey, he observed that musk thistle was under greater insect attack than the other species being studied, so he began to concentrate on that plant and compare his collections with the earlier collections of Andres that had already been identified by USDA taxonomists at the U.S. National Museum. When the comparison was made, Dr. Frick promptly discarded Larinus cynarae F. as a candidate, but Larinus jacae F. was still kept as a possibility. Dr. Frick also took Rhinocyllus conicus (Froel.) adults from milk, musk, and Italian thistles, but found eggs only on milk and musk thistles. He also found Lixus scolopax Boh. on milk, Italian and musk thistles, but recorded feeding and oviposition only on musk thistle. He also encountered three other species of Lixus; Lixus junci Boh. was known to be oligophagous so was discarded, but Lixus elongatus Goeze and Lixus cardui Ol. were at this time still considered as potential candidates. On musk, Italian, and slenderflower thistles, Frick found the weevils Ceuthorrhynchidius horridus and Ceuthorhynchus trimaculatus on the plants and Apion sp. in the crowns. He also found a small black cerambycid associated with the thistles Silybum marianum and a species of Onopordum.

Wanting to select candidate insects for testing and introduction from this list, Dr. Frick then made a study of the insects in the United States that were associated with weedy thistles and found that a trypetid fly, Urophora solstitialis; a tortoise beetle, Cassida rubigionosa L.; and Cleonis piger L., whose common name in Italian translates as artichoke root and crown borer, were known to exist in America. So these insects and the weevil Cyphocleonus tigris Panz., which is known from six widely separated genera of Compositae, including cultivated chrysanthemums, were dropped from further consideration. In the interest of trying to find which insects were actually natural enemies of, or threatening to, artichoke (the closest cultivated relative of the weedy thistles) and thus narrow the list of candidates, Dr. Frick made a most interesting study. In the fall of the year, he collected a large number of artichoke heads that remained in the field after the harvest, caged them, and recorded the insects that were reared from them. Fifteen species of insects emerged. Nine of these had been recorded on the weedy thistles, so this quickly cut by nine the list from which the candidates were to be chosen.

The weevil genus Larinus interested both Frick and Andres because of its ability to inflict severe damage on the weedy thistles, and they hoped to find a really efficient natural enemy. In fact, 18 of the 19 species known in Europe are associated with the composite tribe Cynareae. Right off, they found that four were notorious pests of artichoke or safflower, another five were associated with thistles that do not occur or are only minor problems in the U.S., and three of the remaining nine species were reported only from Centaurea spp., so these insects were not considered further. Of the six left, one was common on Canada thistle; another one, L. turbinatus Gyll., had been collected from five species of Cirsium as well as Carduus, Onopordum, and Galactites; and still another, Larinus sturnus Schal., was known to feed on

artichoke. There was one species not well accounted for, leaving only two, Larinus jacae F. and Larinus carlinae Ol., as possible candidates.

Not all the insects encountered were beetles. There was some initial interest in a trypetid fly, Acanthophilus helianthi (Rossi), but it was later reared from five genera of thistles. Literature records list it from another 13 species. This fly was dropped from further consideration, but not an interest in the trypetids. In addition, Dr. Frick had collected the literature on insects occurring on thistles in Italy and extracted a list of about 50 agricultural pests. This list was most useful to eliminate the testing of any insects that were known as potential pests of useful plants.

While this activity on thistles (which was sandwiched among 10 other weed projects at Rome) was going on, research that was to have a major impact on the biological control of Carduus thistles, especially musk thistle, was occurring to the north. The Canadian musk thistle program as we know it today started as one facet of a broad biological control study conceived in 1961 by Dr. Peter Harris, Canada Department of Agriculture, who at that time was located at Belleville, Ontario. Dr. Harris reasoned that, since many of Canada's worst weeds were in the family Compositae and more specifically in the tribe Cynareae, it would be useful to survey the European insects associated with that tribe. Thus, the Commonwealth Institute of Biological Control Laboratory at Delemont, Switzerland, was contracted to make such a survey, and Dr. Helmut Zwölfer was the project leader.

There were two goals for the survey. One was to obtain as complete a list as possible of insects associated with the thistles in this tribe, and the second was to delineate the host ranges of insects discovered. By arranging the insects into groups according to the plants they attack, it became relatively clear which insects were polyphagous, oligophagous, and monophagous in their feeding habits. With this information in hand, the selection of a list of candidates against any of the weeds in the Cynareae was greatly simplified.

Zwölfer's survey area extended across Europe from western France to eastern Austria, and was bounded on the north by southern Germany and the south by northern Italy. Twelve hundred samples, from 38 species of the subtribe Carduinae, were collected, and when the survey ended in 1964, preliminary screening of some 30 species of phytophagous insects had been started (Zwölfer 1965).

In early 1964 a meeting was arranged between Zwölfer, Andres, and Frick. As one would suspect, a work of the magnitude of Zwölfer's study had a considerable impact on the biological control of thistles in general and musk thistle in particular. A closer liaison developed between Canada and the United States, both on this side of the ocean and through the Canadian-supported projects at the CIBC Laboratory in Switzerland and the Rome laboratory of the USDA.

It didn't take long before a sort of division of labor began between these two similarly oriented facilities, Canada and the CIBC accepting the responsibility for work on some of the insects of common interest, especially those that occurred within working distance of the Swiss laboratory, and the Rome laboratory picking up work on other insects of common interest that occur in Italy. The Canadians first focused on the weevil Rhinocyllus conicus (Froel.) as a candidate insect for musk thistle control. This insect was investigated through 1966, and in 1967 Zwölfer and Harris presented a major report on its

life history, distribution, and host range to support the petition for introduction and release of the weevil into Canada.

In 1968, Rhinocyllus conicus from the Alsace region of eastern France (in the Rhine Valley, near the French, German, and Swiss borders) was released at two places in Canada. One release site was near Regina, Saskatchewan, where 400 adults were released on musk thistle and the other was near Belleville, Ontario, where 370 insects were released on Carduus acanthoides L. Recoveries were made in 1969 but the results were not spectacular, probably because the insects were out of synchrony with the Canadian season (Harris and Zwölfer 1971). Later, in 1971, Zwölfer (1973) presented a paper discussing the insects associated with the heads of musk thistle and suggested a sequence for their most successful introduction.

During the 1964-65 period at Rome (Andres had returned to the USDA laboratory at Albany, Calif.), Dr. Frick continued to synthesize his observations with Andres' earlier, more random field observations of insects associated with musk thistle, and his in-depth literature study. Thus, his choice of candidates began to emerge and lean toward the crown-feeding weevils Ceuthorrhynchidius horridus Panz. and Ceutorhynchus trimaculatus; the flea beetle Psylliodes chalcomera Illig.; and a phytophagous stem-mining syrphid fly, Cheilosia sp. Testing of these species was delayed when Dr. Frick returned to Albany and I was transferred to Rome in 1965. My initial assignment was a survey of the natural enemies of cruciferous weeds. In early 1966 I also picked up the musk thistle work and started host specificity testing of Psylliodes chalcomera, because it was the most abundant of the candidates suggested by Frick. Testing started in April that year and continued through 1967 and 1968. Since the survey on crucifers showed only limited promise, the crucifer work was set aside and my primary attention was devoted to the work on musk thistle. During the collection, observation, and testing of Psylliodes, we discovered that there were normally substantial populations of C. horridus and C. trimaculatus available if one looked in the right place at the right time, so during the first quarter of 1969, host specificity testing was started on these insects as well.

Also during this period, Dr. R. D. Hendrick, then of the Department of Entomology of the Virginia Polytechnic Institute, came to Rome to gain firsthand knowledge of the possibilities of biological control of musk thistle, that plant being recognized as a serious pest in Virginia. Before he left Rome to return to Virginia, it was mutually agreed that he would put emphasis on testing Ceuthorrhynchidius horridus for specificity and that Antonio Rizza and I would finish the testing of Psylliodes chalcomera and Ceuthorhynchus trimaculatus at Rome.

On his return trip to Virginia, Dr. Hendrick visited laboratories in Pakistan, India, and Japan. He encountered two promising species of trypetids in Pakistan, Tephritis heiseri Frnfld. and Terellia serratule (L.), and another two in Japan that are in part responsible for the musk thistle control in that country.

Later that year the release of R. conicus in the United States was approved, and on May 18, 1400 adults were sent to Dr. Hendrick from Rome. Another 2700 were sent to the USDA laboratory in Albany where they were held for observation and further study. Also, Hendrick received two shipments of R. conicus from the Rhine Valley in France in April and July.

Meanwhile, the work continued on the testing of P. chalcomera and C. trimaculatus at Rome, with shipments of these to Albany and of C. horridus

to Blacksburg for continued testing in quarantine. Additional R. conicus shipments were made to Albany for transshipment to Nebraska, where unseasonally dry weather and an inadvertent cutting of the release plot had a serious negative effect on the release, and to Montana, which also received shipments of some R. conicus from musk thistle near Colmar, France.

In 1970, Dr. Ken Frick wrote a petition for the introduction of C. horridus and C. trimaculatus into quarantine in the United States for final testing, and December 15 of that year, 198 C. trimaculatus adults were sent to Albany to be studied in quarantine, and 163 C. horridus adults were sent to Dr. Hendrick at Blacksburg, Va., where an approved quarantine facility had been constructed. Early in 1971 we made a second shipment of C. horridus from Rome to Blacksburg and stopped all laboratory investigations of this insect, but kept on with the P. chalcomera and C. trimaculatus testing. Dr. Kok will discuss what has happened during the post-release period in Virginia, and Norm Rees will tell about the Montana releases.

Returning to the other three insects, Dr. L. T. Kok (who replaced Dr. Hendrick at VPI) completed testing of Ceuthorrhynchidius horridus, and in 1976 this insect was released in Virginia by Kok's group and at Regina, Saskatchewan, by Peter Harris. Because of the importance of artichoke cultivation in California, we at Albany felt that further testing was in order to make additionally sure there was no danger to this crop, valued at about $2,000,000 per year.

Paul Boldt and Antonio Rizza of the Rome laboratory have made additional field cage studies in Italy, and the results of these studies are being weighed prior to deciding whether to release the insect in the West or not, the problem being the adult's ability to feed and oviposit on artichoke in laboratory tests despite its failure to attack artichoke in Italy.

Following the clearance of C. horridus, the testing of Ceutorhynchus trimaculatus was assumed by Dr. Kok in cooperation with the USDA. To date, there have been no surprises emanating from this testing, and the insect is expected to be released in 1978 or 1979.

In recounting this history there are several loose ends that remain to be picked up. One of these is the tortoise beetle Cassida rubigionosa L. This insect, already found in North America, was screened by Zwölfer in 1963 and found to feed on artichoke under caged conditions. This, plus the fact that the insect is heavily parasitized in its eastern range, eliminated serious interest in further study.

Another loose end is the flea beetle Psylliodes chalcomera. High hopes were held for this insect, which is quite damaging to musk thistle and has never been recorded as an artichoke pest throughout its wide distribution range in Europe. It was studied intensively, and all uncaged tests indicated that it posed no danger to artichokes, but when caged with artichoke as the only food source, a small percentage of the larvae can complete their life cycle on this plant (P. Boldt and P. H. Dunn, unpublished data). This caged feeding behavior indicates that artichoke is not a good host, but since it is nontoxic to the insects, they can sustain themselves on it. Apparently, the mechanism that protects artichoke from P. chalcomera in uncaged trials breaks down in the test cages. While this is a real barrier to insect attack in the field and accounts for the fact that it is not a pest of artichoke, the mechanism is not understood. Thus, P. chalcomera cannot be released until we understand precisely how the beetle selects its host plant or until an experiment can be designed to show that feeding on artichoke in cage trials is a behavioral artifact of caging the insect and not its true behavior. While pondering

this problem, we have put Psylliodes on the back burner and are going ahead with the less problematical ceutorhynchid weevils. Paul Boldt, who transferred to Rome following my return to the United States, has taken over these studies and initiated work on the phytophagous syrphid flies in the genus Cheilosia. According to Dr. Frick's original list, these offer good possibilities for thistle control, but that's future history. One of Boldt's first contributions in his new position was to bring to attention an Australian publication which pointed out that our North American musk thistle isn't really Carduus nutans but rather C. thoermeri Weinm. (Doing et al, 1969).

Through a rather complicated series of events Dr. M. K. McCarty became involved in the biological control of musk thistle and in 1976 went from Lincoln, Nebraska, to Peshawar, Pakistan, to visit Dr. S. M. A. Kazmi, who revised the genus Carduus. The purpose of this trip was to bring order to North American musk thistle taxonomy so we would know which was our actual target plant. This important facet of our biological control program has moved to the final developmental stages, so it is evident that the last chapter of the history of the biological control of musk thistle hasn't even been outlined much less written.

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THE GENUS *CARDUUS* IN THE UNITED STATES[1]

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Many pests that become serious problems are introduced from abroad. This is true of musk thistle, currently known as Carduus nutans L. Seldom are the date and method of entry of an alien organism recorded. As plant sciences were not well developed early in our nation's history, it is not surprising that there are scant records as to the date and location of the first Carduus plants in the United States. Apparently, our earliest records are from Pennsylvania a little over 100 years ago (Stuckey and Forsyth, 1971). More samples were recorded near the turn of the century. At this time Carduus plants were only a taxonomic oddity and of little or no concern. No mention was made of these thistles in the early 1900s as a weed pest.

By the early 1940s musk thistle began to be mentioned as a potential weed problem. By the mid-1940s, following World War II, when some of our modern pesticides surfaced--such as the phenoxy herbicides and the chlorinated hydrocarbon insecticides--organized pest control efforts were started. Some of the early work on musk thistle, in addition to that at Nebraska, was in Kentucky (Freeman, 1960).

About this time several states enacted noxious-weed-and-seed laws requiring certain weeds to be controlled and limiting movement of crop seed contaminated with these weed seed. Kansas and Nebraska, probably the areas with the largest and most severe infestations of musk thistle, became aware of the problem and its seriousness. Nebraska's first law was enacted in 1939 and expanded in 1945. Musk thistle was added to its noxious-weed list in 1959 and plumeless thistle (Carduus acanthoides L.) in 1965. Kansas put them on its weed list at about the same time. Programs of control in these two states soon became multimillion dollar enterprises.

Biological control research efforts in Europe had revealed a number of insects thought to hold promise for use on Carduus species (Zwölfer, 1965, 1967). One, Rhinocyllus conicus (Froel.), was cleared for release in North America in the late 1960s. Early releases of Rhinocyllus were made in Canada in 1968 (Harris and Zwölfer, 1971) and in Montana (Hodgson and Rees, 1976), Virginia (Surles et al., 1974), and Nebraska in 1969. Establishment occurred in Canada, Montana (Rees, 1977), and Virginia (Kok, 1974). The Nebraska releases were lost and work was not resumed until 1972. In 1974, extremely high temperatures, low humidities, and above-normal wind velocities in Nebraska release areas dried the thistle heads, and a considerable number of weevil larvae and pupae perished.

In 1975, weevils were collected in Montana with the hope that the successful colony there might have made some adaptive changes that would improve the chances for better colonization in Nebraska. When the shipment of weevils and some thistle material used for packing, food, and moisture arrived, I found that the thistle was different from that found in Nebraska.

I had seen thousands of acres of thistles in Kansas, Nebraska, and surrounding states as well as mounted specimens from Illinois, Wisconsin, Minnesota, Kentucky, and Oklahoma. All of this plant material appeared to be essentially the same and at this point I felt no reason to question its classification, acknowledging that variation in site, fertility, moisture, etc. can produce differences in appearance.

The differences observed between the plant material from Montana and that in the Plains area were not described in available manuals. In conference with personnel of the Biological Control of Weeds Laboratories at Albany, California, and Rome, Italy, it was decided that an effort should be made to resolve the identity problem. With the help of personnel from the Rome Laboratory, a schedule was devised for me to work with Dr. S. M. A. Kazmi, the author of a monograph on Carduus, and to make a collection trip with Mr. Paul E. Boldt of the Rome Laboratory on a trip through known Carduus source regions to collect plants and associated insects. This we did from June 19 to July 12, 1976.

The materials were collected in triplicate. One set was retained at the Rome Laboratory, one set was left at the Botanic Institute at Munich for Dr. Kazmi, and one set returned with me to Lincoln. These plants were identified and determinations corroborated by Dr. Kazmi late in 1976. Since that time, I have been collecting materials and have started a nursery at Lincoln. Collections from major herbaria in the United States, including the Gray Herbarium, New York Botanical Garden, Missouri Botanical Garden, and others, have been assembled. Over 500 pressed specimens have been examined, and what appeared at first to be a difficult but manageable task has become very complicated.

The original three species of Carduus listed in the standard manual, "Gray's Manual of Botany" (Fernald, 1950), have turned into at least six, possibly seven, species. Numerous other bits of information have surfaced, such as the slender-flowered, or Italian, thistles, thought to be restricted to the West Coast, actually were collected in Eastern states before the turn of the century.

The three species of Carduus listed in Gray's Manual are nutans, crispus, and acanthoides, with a brief key that can be simplified as follows:

Heads nodding, solitary, large. . . . . C. nutans
Heads ascending, often clustered, small
  Involucre hemispherical, 1.5-2.5 cm broad,
    outer phyllaries somewhat herbacious and
    spreading . . . . C. acanthoides
  Involucre ovoid, 1-1.3 cm broad, outer
    phyllaries rigid, hardly spreading . . . . C. crispus

For many years this was accepted as the definitive document for species identification. This made no allowance for either the slender-flowered thistles or the complex of species in the large-flowered group. Thus, when Dr. Hodgson at Bozeman sent a specimen to Beltsville for identification, it was called C. nutans. The material we have in the Great Plains was also designated C. nutans, as the document set up as official contained no other descriptions.

Recently, "The Thistles of Canada" was published (Moore and Frankton, 1974). In this manual the initial breakdown to species was the same as that in Gray's Manual above, i.e., C. nutans, acanthoides, and crispus. However, they broke out three subspecies of nutans as ssp. nutans, ssp. leiophyllus, and ssp. macrolepis. Moore and Mulligan (1956) described a hybrid of nutans and acanthoides that they called C. x orthocephalus. There were apparently large populations of this hybrid in Gray County, Ontario, and they have published some detailed studies of this population (Moore and Mulligan, 1964).

It becomes obvious that there are questions about the proper classification of the main body of Carduus plants that exist in the United States. Doing et al. (1969), in studies in Australia, called the plants they were using members of the C. nutans group (nutantes of Kazmi, 1964a). They further determined that portions of their populations were C. nutans and portions C. thoermeri. They also said that plants raised from seed from Argentina and the United States were C. thoermeri.

The Flora Europaea, 1976 edition, in its treatment of Carduus does not initially key out any of the species of the nutans group but lumps them in a group of eight (Tutin et al., 1976). It then describes the group as follows: "A difficult group in need of further study. Though a few extreme taxa are easily recognized, there is considerable variation in hairiness, leaf-size, spine-length, peduncle-diameter, width and shape of bracts, and corolla-length. This variation is almost continuous and intermediates between taxa can be found." It then goes on to key out these eight species and list three subspecies each of nutans and macrocephalus.

I have tried to use information from Kazmi, the Flora Europaea, a treatment of Carduus in Germany by Kazmi (1964b), and excerpts from a flora of the Ukraine to study the plant materials on hand. From this I believe that we have perhaps as many as seven species of Carduus in the United States and that most Carduus thistles are C. thoermeri. Some form of nutans probably exists, as well as macrocephalus in the large-flowered group, crispus and acanthoides in the small-flowered group, and pycnocephalus and tenuiflorus in the slender-flowered, or Italian, thistle group.

The present collections of plant material at Lincoln will be enlarged. The nursery work involving plants from many locations in the United States growing under one set of site conditions will be continued and expanded. This will allow comparisons of plant materials with a minimum of variability due to edaphic and climatic factors. The plant material studies are being supported by cytological examination. The goal is to prepare a manual that will assist in the identification of the Carduus species that exist in the United States at this time.

[1]Cooperative investigations of the USDA Science and Education Administration and the Nebr. Agr. Exp. Sta.

[2] Personal study of specimens of Carduus from the following herbaria: Missouri Botanical Garden, St. Louis, Missouri; Field Museum of Natural History, Chicago, Illinois; Gray Herbarium of Harvard University; University of California, Davis; University of Wisconsin; University of Kentucky; Duke University; Virginia Polytechnic Institute; and Iowa State University.

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FOREIGN EXPLORATION FOR THE BIOLOGICAL CONTROL OF CARDUUS SPP.

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Several species of Carduus, introduced into North America probably from Europe, severely infest pastures and croplands in 12% of the counties of the United States (Dunn 1976). Control of these thistles through the release of imported natural enemies promises to be successful because the plants are of foreign origin and have few natural enemies in North America. Despite wide abundance and severeness, control can be directed against future rather than present populations of Carduus (Frick 1974, Goeden 1977).

Once importation of biotic agents was selected as a method of control for Carduus, the following steps were necessary: (1) study of the plant and a survey of phytophagous insects associated with it in the problem area, (2) foreign exploration--a companion study of the plant and its associated insect species in the area of the plant's origin, (3) detailed host plant specificity tests on candidate biocontrol agents, with detailed biology studies being continued in the area of origin, and (4) release and evaluation in problem areas (Goeden 1977, Grabau and Spencer 1978). Sometimes these steps are conducted simultaneously, and new information may require that a step be repeated after a number of years. No single part of the program can be neglected, nor is one part any more important than another. Shortcuts or lack of adequate followup studies would deprive researchers of important information necessary for an evaluation of the impact of selected insects in their new environment and for prediction of possible side effects of the proposed introduction.

This paper discusses the foreign exploration aspect of importation of biotic agents against the Carduus nutans L. group. The majority of North American Carduus belong to this group. The biotic agents of C. pycnocephalus have previously been reported by Goeden (1974) and Zwölfer (1965).

Three phases of foreign exploration are: (1) a study of the host plant (weed), (2) a survey of natural enemies associated with the weed, and (3) a selection of biological control candidates from among these enemies.

The first phase includes the determination of the center of origin of the target plant, as well as knowledge of its various forms and habitats. The problem was compounded in the case of musk thistle because of conflicting taxonomic descriptions of the species of Carduus in Europe, which gave the false assumption for many years that only one plant species, C. nutans, was present in the United States. In an attempt to resolve this difficulty, plant specimens collected from the field and specimens from several museums in Europe were studied to determine the amount of variation which existed among plants. Observations were also made on plant distribution and habitat. Correctly identified European specimens were compared with United States specimens.

Southern Europe is considered to be the center of origin for Carduus because of the many endemic Carduus species (Kazmi 1964). Linnaeus first

described C. nutans from Sweden in 1753. Since then, many species and subspecies of plants near C. nutans have been described in other areas by various authors. In Italy, Fiori (1927) called all large-headed plants C. nutans and listed 4 subspecies. Kazmi (1964), on the other hand, described 7 species, of which at least 3 occur in Italy. Some of these species were formerly considered forms of C. nutans.

On the basis of this study, the European distribution of the 4 common large-headed species of Carduus was as follows: (1) C. nutans is found in central and northern Europe and in the Apennine Mountains in Italy. (2) C. macrocephalus Desfontaines is common in the Mediterranean area and abundant in Italy, Yugoslavia, and Greece. (3) C. micropterus (Borbas) Teyber is found in southeastern and north-central Italy. (4) C. thoermeri Weinmann is common in the Balkans from Austria to eastern Turkey. All species have the characteristic constriction of the middle involucral bract, but they can be separated on the basis of the differences in the shape of the bracts, shape and pubescence of the leaves, and their different geographical distribution.

The 3 European species present in the United States and Canada are: (1) C. nutans, distinguished by its long, tapering bracts and palmately cut leaves. (2) C. macrocephalus, distinguished by its long, tapering bracts and pinnately cut leaves, and (3) C. thoermeri, distinguished by its wide, short bracts and wide, hairless, palmately cut leaves (Kazmi 1964).

The second phase of the foreign exploration program was a survey of natural enemies of Carduus which occurred within the area of origin. Previous surveys made in various parts of the world were reviewed to determine the numbers of monophagous or oligophagous insects in each area. The obvious polyphagous species were eliminated as potential biological control agents. Although these may do severe damage, they cannot be relied upon for continuous performance as they may easily select other plants of recognized value for feeding and oviposition.

From a species of large-headed Carduus, 11 endophagous species were recorded from South Dakota (Morihara and Balsbaugh 1976). These species, however, occurred in small numbers, were not specific to Carduus, or were recorded as economic pests, thus confirming the need to explore foreign areas for biocontrol agents. On C. nutans, 32 endophagous species were recorded from central Europe (Zwölfer 1965). On the basis of Zwölfer's survey, Rhinocyllus conicus (Froelich) (Coleoptera: Curculionidae) was selected, tested, and first released in Canada (Zwölfer 1967, Harris and Zwölfer 1971). On C. macrocephalus, 42 endophagous species were recorded from Italy (Boldt, unpublished data). On Carduus edelbergi Rechinger, a related species, 17 endophagous species were recorded from Pakistan (Baloch et al. 1971). The large number of insect species recorded in central and southern Europe, and the similar morphological structures of C. nutans and C. macrocephalus, may indicate a close relationship of the plants. It is probable that these surveys were made near the center of origin of the Carduus group.

Within the scope of the present survey, several other species of Carduus in Italy have been partially evaluated for biocontrol candidates by Andres, Frick, and Dunn (former entomologists of this laboratory), but the main emphasis has been on insects of C. macrocephalus because it is the dominant large-headed Carduus in Italy and is thus available for study.

All species collected on C. macrocephalus were initially evaluated on the basis of their feeding behavior, determined through a literature search and preliminary host testing, and of their observed impact on the plants. Of the 93 insect species collected on Carduus, 44% were polyphagous on plants outside

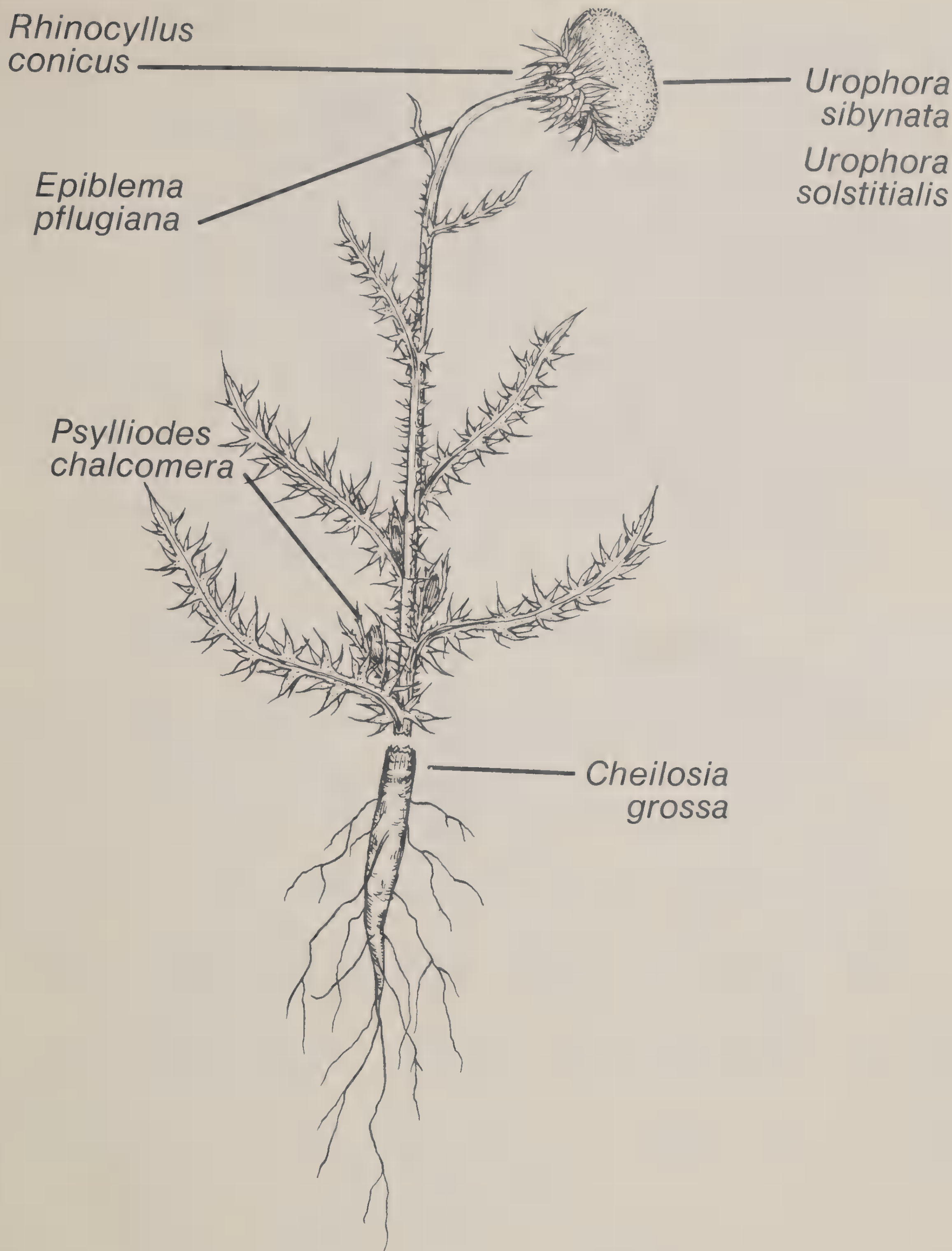

Figure 1.--Potential biological control insects that attack the bolting stem or flower head of Carduus macrocephalus.

the family Compositae, 23% were restricted to feeding on Compositae, 12% were recorded only on the tribe Cynareae, and 21% were recorded only on the subtribe Carduinae. From this last group of 20 insects, 7 have been reported as economic pests, usually on artichoke, Cynara scolymus L., while 4 apparently caused no important damage to that plant. Thus, only 9 of the original 93 insects were considered as possible biocontrol candidates.

The third step in foreign exploration was to use the knowledge gained from the survey to select those natural enemies with the most potential for detailed host specificity testing. Available time, money, and manpower did not permit detailed study of all candidates simultaneously. Therefore, the following questions were asked when making this selection: What part of the plant does the species attack? Does it complement other biocontrol agents and other control methods? Is it host specific?

The insect species ultimately selected were those which attack different parts of the plant or different stages of development. Carduus can be considered as having 2 growth stages; the bolting stem which produces flower heads (fig. 1) and the leafy rosette (fig. 2). Each stage has its own insect complement.

INSECTS WHICH ATTACK THE BOLTING STEM OR FLOWER HEADS

R. conicus (Froelich) (Coleoptera:Curculionidae). - Larvae feed in the immature heads of Carduus. Has been released in Canada, the United States, and New Zealand (Harris and Zwölfer 1971, Surles et al. 1974, Jessep 1975).

Urophora sibynata (Rondani) and U. solstitialis (L.) (Diptera: Tephritidae). - Larvae cause gall formations in the flower head, which becomes lignified. Although capable of severely limiting seed production, the possible competition of Urophora spp. with R. conicus suggests a rather limited potential at this time. If, in the future, a decision is made to test these species, data on plant impact, density, biology, and collecting areas are available.

Cheilosia grossa (Fallen) (Diptera: Syrphidae). - Larvae feed in the stems and roots of musk thistle, retarding growth and causing the production of small heads (Boldt 1978).

Psylliodes chalcomera (Illiger) (Coleoptera: Chrysomelidae). - Larvae cause extensive damage to the leaf and flower buds (Dunn and Rizza 1976). However, this insect was rejected as a biocontrol agent after tests showed that it would attack artichoke (unpublished data).

Epiblema pflugiana (Hayworth) (Lepidoptera: Olethreutidae). - Larvae feed for one generation in the peduncle at the base of the flower head, preventing the head from producing seed. Further studies are planned for E. pflugiana. It has 3 generations per year in the Rome area. The other 2 generations feed in the rosette.

INSECTS WHICH ATTACK THE ROSETTE

Ceuthorrhynchidius horridus (Panzer) (Coleoptera: Curculionidae). - The first insect to be tested and released against the rosette of C. thoermeri in the Eastern United States (Kok et al. 1975) and against C. nutans in Canada (Harris, personal communication). It is being considered for release in New Zealand (Jessep, personal communication) and in the Western United States.

Ceutorhynchus trimaculatus F. (Coleoptera: Curculionidae). - The larvae also feed in the crown but complement C. horridus in Europe by feeding about 1 month later. The larvae of both species are sometimes found in the same plant

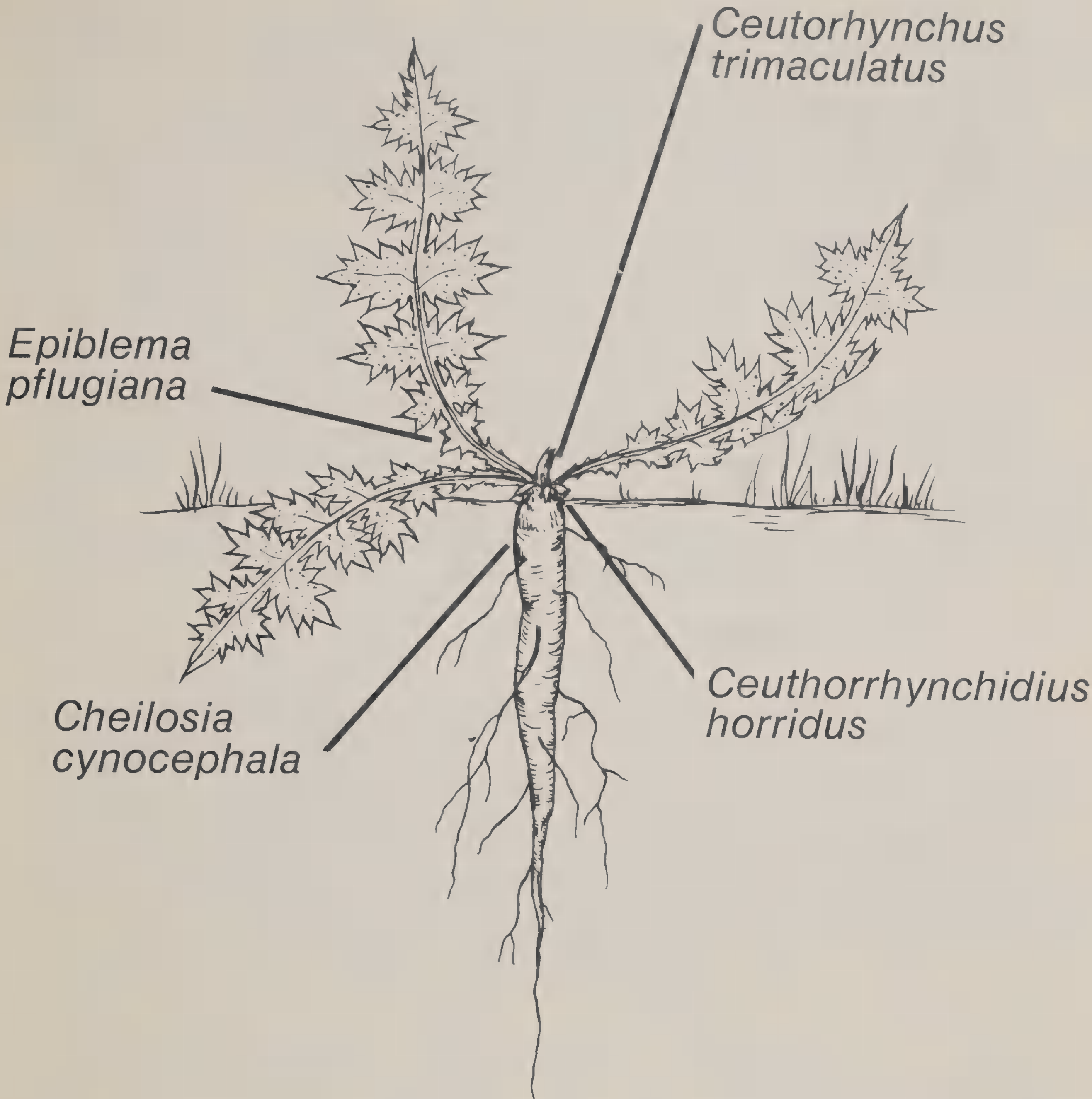

Figure 2.--Potential biological control insects that attack the rosette of Carduus macrocephalus.

but appear compatible, even at high densities. Research leading towards the possible introduction of C. trimaculatus into the United States is currently underway (Boldt 1978).

Epiblema pflugiana (Hayworth) (Lepidoptera: Olethreutidae). - Two generations of the larvae attack the rosette stage of Carduus, feeding in the crown and the upper portion of the roots. Larvae in both these generations are capable of retarding the growth of the plant. Both generations occur before oviposition by C. horridus begins.

Cheilosia cynocephala Loew (Diptera: Syrphidae). - Larvae attack the rosette in the fall by feeding from the outside to the center of the root. It complements C. horridus by feeding below the crown.

PLANT PATHOGENS WHICH ATTACK THE PLANT

In addition to these 9 insects, 2 plant pathogens are also being considered. A rust, Puccinia sp. (Basidiomycetes: Pucciniaceae) attacks leaves of the rosette and the basal part of the plant. An unidentified smut fungus (Basidiomycetes: Ustilaginaceae) attacks late-maturing seed heads. The smut completely eliminated all seed production in heavily infested plants in several areas of Italy. This fungus may be a good complement to R. conicus, which feeds in the early flower heads, because it appears in the summer during the time of seed formation.

SUMMARY

Over a period of 15 years, the Carduus foreign exploration project has gathered information about the plant and its natural enemies. Four species of large headed Carduus in Europe can be distinguished morphologically and geographically. Two of these species have been surveyed for biological control candidates. Of the 93 insect species on C. macrocephalus 9 species have been selected as biocontrol candidates, 2 of which have already been released. Two pathogenic fungi have been found which deserve further consideration. The importation project will continue to offer input toward the objective of reducing Carduus to an acceptable density in North American pasture land.

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CARDUUS THISTLE DISTRIBUTION AND BIOLOGICAL CONTROL IN THE NORTHEASTERN STATES

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The northeastern region includes West Virginia, Maryland, Pennsylvania, New York, Delaware, New Jersey, and the six New England States. *Carduus* species occurring in this region are *C. acanthoides* (plumeless thistle), *C. crispus* (curled or welted thistle), and *C. nutans* or *C. thoermeri* (nodding or musk thistle).

Carduus crispus is an economic problem only in West Virginia, where *Rhinocyllus conicus* has been established by the West Virginia Department of Agriculture in Monroe County for its control (Hacker 1975). This thistle has been present in the general area since about 1920 (Virginia record, Johnson 1974).

Carduus nutans was first recorded in the U.S.A. in 1853 at Harrisburg, Pa. (Stuckey and Forsyth 1971), where it still remains as a serious pest. Other early northeastern introductions were in ballast dumped at Hoboken (1893) and Camden, N.J. (1880), at the harbor of Providence, R.I. (1890), and at Washington, D.C. (1897). *Carduus acanthoides* similarly appeared first at ballast dumps at Camden, N.J. (1878), and Hoboken, N.J. (1880), and at the Providence, R.I., harbor (1893); it was found in Ohio in 1878.

Carduus nutans and *C. acanthoides* in the northeast often occupy the same habitats, such as overgrazed pastures and roadsides, sometimes occurring as mixed stands. These plants are troublesome primarily in the Great Valley region, which is a northeastward extension of the Shenandoah Valley of Virginia and located between two long, parallel mountain ridges. Serious infestations are found from northern West Virginia to near Hagerstown and Frederick, Md., to Harrisburg, Pa., and eastward past Allentown into western New Jersey (Fig. 1). As noted in Ohio by Stuckey and Forsyth (1971) and in Virginia by Hensley (1973), these thistles in the northeast create economic infestations primarily when growing in shallow soil over limestone. Scattered plants or patches may be found elsewhere, and due to their conspicuousness these may be relatively well represented in herbaria. However, the early introductions outside the limestone zone apparently did not create major infestations there (see Fig. 1).

For example, as one progresses from east to west across the State of Maryland, four zones are encountered:

(1) Eastern Shore (Delmarva Peninsula) - sandy soils.

(2) Piedmont Plateau (area around Washington, D.C.) - primarily poor red clay or mica-schist soils.

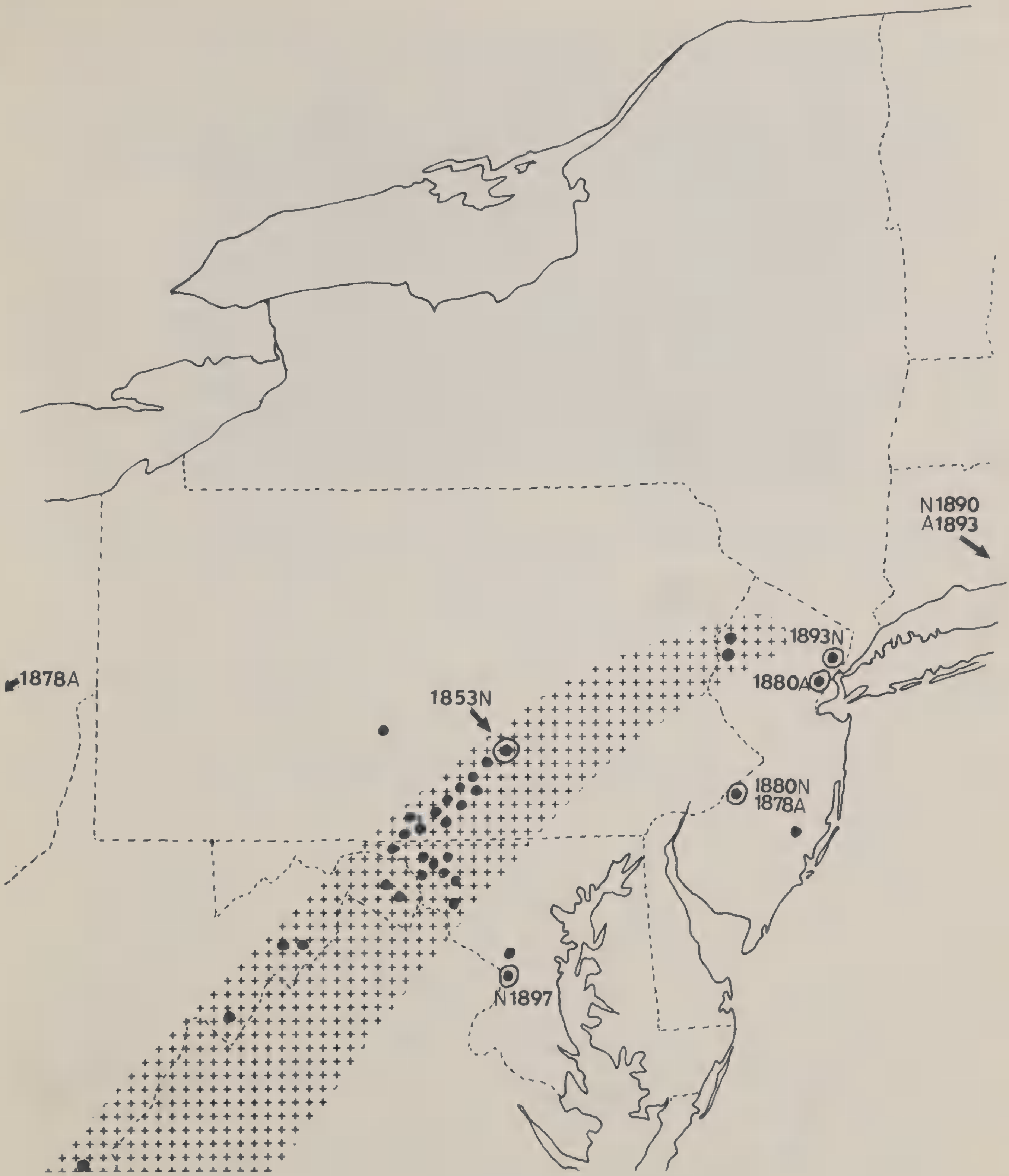

Figure 1.--General distribution of economic infestations of Carduus spp. in the northeast (stippled area). Sites of early introductions are indicated by circled dots with dates (N = C. nutans; A = C. acanthoides). Areas where Rhinocyllus conicus has been released are indicated by dots.

(3) Great Valley - well-drained fertile soils developed over limestone.
(4) Western Maryland beyond Hancock - shale, sandstone, or schist.

Bull thistle (Cirsium vulgare) is the predominant thistle in the coastal eastern region (zones 1 and 2) on the red clay or sandy soils; Carduus thistles occupy the same ecological niche in zone 3 (limestone soils), and Canada thistle (Cirsium arvense) becomes predominant in western Maryland as well as in other areas north and west of the Great Valley.

The distribution of Carduus acanthoides and C. nutans is positively correlated with the location of fertile soils developed over limestone in three contiguous counties, Franklin, in Pennsylvania, and Frederick and Washington, in Maryland. In Franklin County (Long 1975), this valley soil, referred to as the Hagerstown-Duffield association, occupies 32% of the county, where nearly all is cleared for crops, orchards, hay, and pasture. Carduus thistles in Franklin County also extend their range to somewhat overlap adjacent areas of thicker valley soils overlying limestone (Murrill-Laiding formation) or shale and sandstone (Weikert-Becks-Bedington association). The reddish to yellowish soils of the Hagerstown-Duffield-Frankston association occupy 46% of the total area of Washington County, Md. Although cultivation may be hindered by the numerous limestone ledge outcrops, this most fertile soil in the county provides high yields in corn, small grains, hay, and pasture (Matthews 1962). About 10% of Frederick County is occupied by the well-drained Duffield-Hagerstown-Sasquatchie-Athol valley soils developed from limestone and shale over limestone rocks much less acidic than surrounding areas. Although there may be massive outcrops of hard limestone, excellent yields support numerous farms and productive dairying. Although Carduus is most prevalent on these soils, some thistles grow in adjacent poorer mica-schist-derived soils of the Piedmont Plateau where underlain by marble or limestone. These plants are rarely found to the west or north on stony, steep mountain soils or on the shallow soils of red shale and sandstone in the valleys. As may be seen from the above data, Carduus thistles are a particularly serious economic pest because they interfere with agriculture on the most productive soils of the region (Fig. 2.).

The geographic distribution of economic infestations of Carduus thistles in the northeast appears to be long-standing and related to the availability of soils formed over limestone. This does not seem to be the case with the recent and rapidly spreading infestations occurring in the central Midwestern and Mountain States in various soils (McCarty 1964; McCarty et al. 1973; Dunn 1976). According to Doing et al. (1969), C. nutans in Australia grows in moist, neutral, well-drained soils over basalt or granite, fertile, calcareous soils being rare in climatic zones suitable for this plant.

The USDA biological control program for Carduus nutans and C. acanthoides in the northeast began in July 1975 with the release of Rhinocyllus conicus (Froelich), collected from a large population established in northern Virginia by L. T. Kok. Releases were as follows:

(1) 300 weevils at 4 locations in Maryland.
(2) 600 at 2 locations in New Jersey.
(3) 200 at 6 locations in central Pennsylvania.

Figure 2.--Dairy cattle in a typical overgrazed pasture infested by C. nutans in Pennsylvania. Not only are the thistles not eaten, but the cattle do not reach for the edible plants growing near them.

In 1976, May and June releases, expected to be more effective (Kok, 1974), were as follows:

(1) 420 weevils at 10 locations in Maryland.
(2) 300 weevils at 8 locations in Pennsylvania.

In 1977, spring releases continued as follows:

(1) 350 weevils at 2 locations in Maryland.
(2) 890 weevils at 7 locations in Pennsylvania.

Initial establishment at the 1975 release site at Frederick, Md., was indicated by a 4% thistle-head infestation rate in 1977. There is no evidence yet that other populations are established, and monitoring continues.

These releases of R. conicus were made in heavily infested pastures, in vacant land, and also along highways where Carduus thistles have crowded out crown vetch (Coronilla varia) planted on steep banks for erosion control. In addition, releases were made at research plots prepared at Beltsville and Frederick, Md. Many releases were made in cooperation with officials of the Maryland State Highway Administration, the Pennsylvania Department of Transportation, and the Pennsylvania and Maryland State Departments of Agriculture. In addition, R. conicus was provided for release by the New Jersey Department of Agriculture. Counties in the northeastern region where R. conicus has been released are: Prince George's, Washington, and Frederick (Maryland); Franklin, Cumberland, and Centre (Pennsylvania); Hunterdon, Warren, and Burlington (New Jersey); and Monroe (established), Jefferson, Berkeley, Pendleton, Grant, and Hardy (West Virginia, Hacker, 1975; Moore, 1976).

Cassida rubiginosa, an accidentally introduced European chrysomelid, is abundant on C. nutans and C. acanthoides in Maryland and Pennsylvania. This defoliator, which also attacks Canada thistle, Cirsium arvense (L.) Scop., (Ward and Pienkowski 1975) does not seem to significantly reduce the vigor of Carduus thistles, although leaves may be extensively damaged in some areas.

When sufficient insects become available at the first United States establishment sites in Virginia (Kok and Trumble 1977), the European thistle rosette weevil, Ceuthorrhynchidius horridus, will be collected and distributed in the northeastern States to augment R. conicus. As soon as additional biological control organisms are tested and approved, they will also be distributed and released in the northeastern States (see Batra et al. in preparation).

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STATUS OF BIOLOGICAL CONTROL OF MUSK THISTLE IN VIRGINIA

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Musk thistle (*Carduus nutans* L.)[1] and plumeless thistle (*Carduus acanthoides* L.) are introduced plants which are generally recognized as serious weed problems in pastures, ranges and croplands in many parts of the U.S.A. In the absence of their natural enemies, the thistles have spread rapidly. Musk thistle is now present in more than half of the states in the country, many of which consider it to be a noxious weed (Dunn 1976). In Virginia alone, over 150,000 acres of pastures are very heavily infested (Kates *et al.* 1972); the total acreage with thistle infestation is substantially greater. Distribution of the thistles is concentrated along the northeast-southwest third of the state (Fig. 1). Musk thistle predominates around the northeast corner and the middle third of the "thistle belt"; the rest is dominated by plumeless thistle. The infested area is increasing despite more than a decade of chemical control by individual farmers and by the Virginia Department of Agriculture and Commerce. This is largely due to the prolific reproductive capacity of the thistles; estimates range from an average of 11,000 seeds per plant (Feldman *et al.* 1968) to as many as 120,000 (Lacefield and Gray 1970). The seeds generally have a high rate of germination spread over a number of years. Although timely applications of herbicides provide temporary relief, they are increasingly expensive and do not provide adequate control because of the variation in germination rate of the plants. In addition, thistles on marginal land are often left untreated and are continuous sources for reseeding. There is currently no efficient, economical and environmentally acceptable method for large-scale control of either musk or plumeless thistle. The ultimate objective in our research is to provide an effective alternative to the heavy dependence on herbicides in thistle control.

The first concerted attempt at biological control of musk thistle in Virginia was initiated in 1969 with the introduction of *Rhinocyllus conicus* (Froel.). This was subsequently followed by studies on three other insects: *Ceuthorrhynchidius horridus*[2] (Panzer), *Ceutorhynchus trimaculatus* (Fabricius) and *Cassida rubiginosa* Müller (Table 1). *R. conicus*, the thistle head weevil, was imported from France and released on the basis of specificity tests by Zwölfer (1967) which showed that it would not adversely affect beneficial plants. Eggs of this weevil are laid on the bracts of thistle heads and the larvae feed on developing seeds within the receptacle. It is successfully established in several states in the U.S.A. and Canada (Harris and Zwölfer 1971; Hawkes *et al.* 1972; Surles *et al.* 1974; Goeden and Ricker 1974; Kok 1974 & 1977; Rees 1977). Our research on *R. conicus* can be divided into 2 phases: (i) introduction and releases of imported adults in 1969/70 and (ii) evaluation of releases and establishment of weevils relocated from established sites since

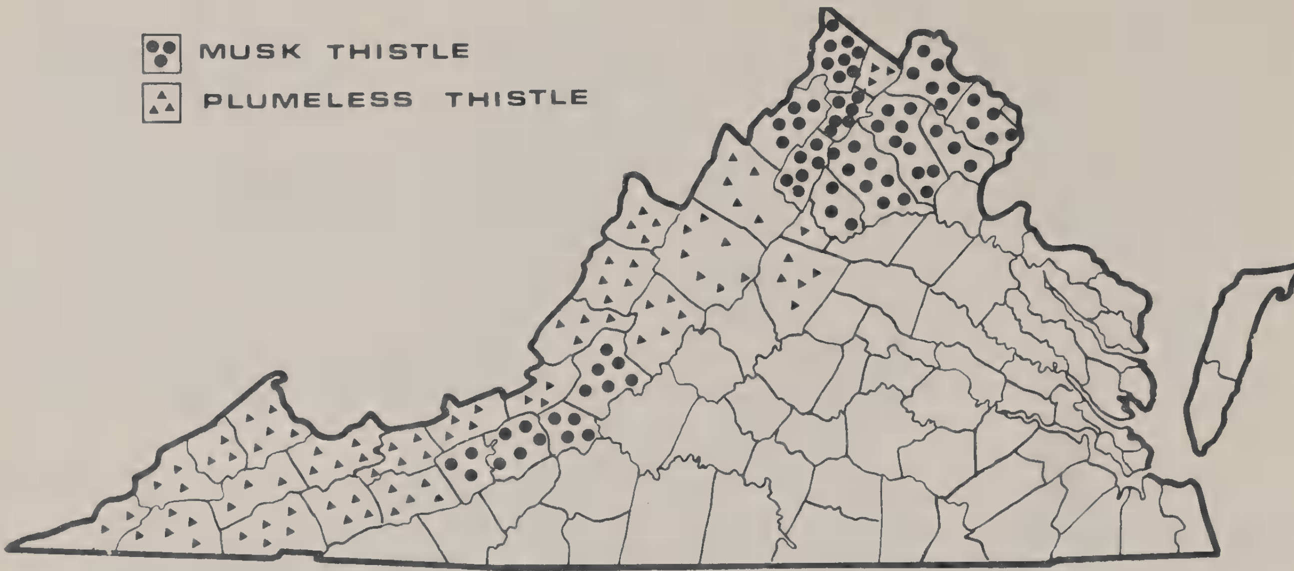

Figure 1.--Distribution of *Carduus* thistles in Virginia.

TABLE 1.--Biological control of *Carduus* thistles in Virginia

Biocontrol agent	Source	Damage on thistles
1. *Rhinocyllus conicus*	France	Head-feeder
2. *Ceuthorrhynchidius horridus*	Italy	Rosette-feeder
3. *Ceutorhynchus trimaculatus*	Italy	Rosette-feeder
4. *Cassida rubiginosa*	Northern Virginia	Leaf-feeder

1-3 = Curculionidae; 4 = Chrysomelidae

TABLE 2.--*R. conicus* on musk thistle in Virginia

Date/Phase of work	# release sites	# sites with establishment	Current control status
1969/70			
Introduction & release	8	4	3 substantial* 1 partial
1972/74			
Relocation from established sites	12	12	3 substantial* 9 partial

*Over 75% reduction in thistle density.

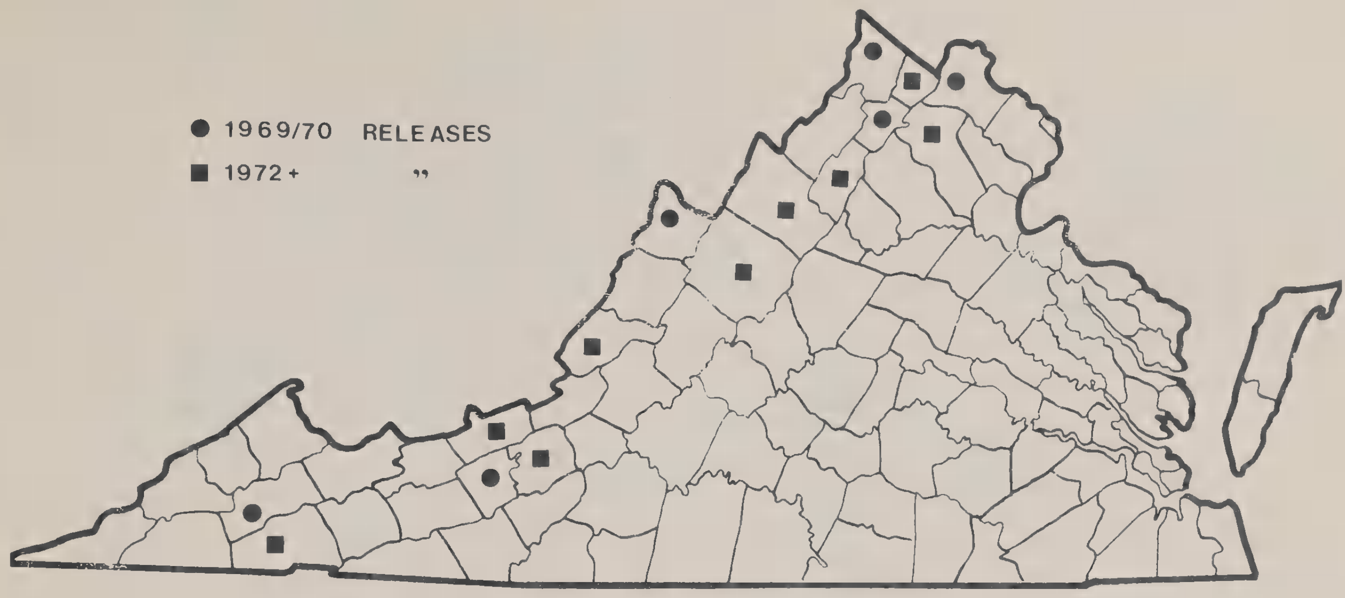

Figure 2.--Established sites of *R. conicus* in Virginia.

1972. The current status of 20 releases which have been monitored annually is summarized in Table 2. Four of 8 releases made in 1969/70 resulted in successful establishment and 3 have shown substantial control. Of the 1972/74 releases, all 12 became established, and 3 have shown substantial reduction in thistle density. The results confirmed that spring releases of reproductive adults were superior to summer releases for establishment (Kok 1974). The potential of *R. conicus* for control of musk thistle was first shown by the reduction of this thistle at one of the releases in Pulaski County (Kok and Surles 1975). To date, 6 experimental sites have shown reductions of at least 75% of the original thistle density. The evidence shows that it takes 5-6 years for an established population of *R. conicus* to have a significant impact on musk thistle. This has been achieved despite the adaptation of some parasites to the larval stage (Surles 1974; Surles *et al.* 1975), and the winter mortality of adult weevils (Kok 1976). The weevil's success can be attributed in part to its ovipositional preference (Surles and Kok 1977) and good synchronization with the musk thistle (Surles and Kok 1976). Wide dispersal of the weevil in Virginia has occurred in 3 counties: Montgomery, Pulaski and Warren County. It has been recovered from at least 15 counties (Fig. 2).

Investigations on the second weevil, *C. horridus*, are summarized in Table 3. This rosette weevil was imported from Italy under quarantine for further host specificity testing on the basis of Frick's (1969) recommendation. It was approved for field release in Virginia in June 1974, after undergoing three years of intensive host specificity testing (Ward *et al.* 1974; Kok 1975a). The larvae damage thistle rosettes by feeding on the meristematic tissues. Biological studies (Kok *et al.* 1975; Ward and Kok 1975) have indicated its potential in the control of both musk and plumeless thistle. Between 1974-76, there were 4 releases on musk thistle in Montgomery and Pulaski County (Fig. 3) (Table 4). The first release consisted of 30 adults and 2,000 first instars on musk thistle in Montgomery County in October 1974. The adults were placed

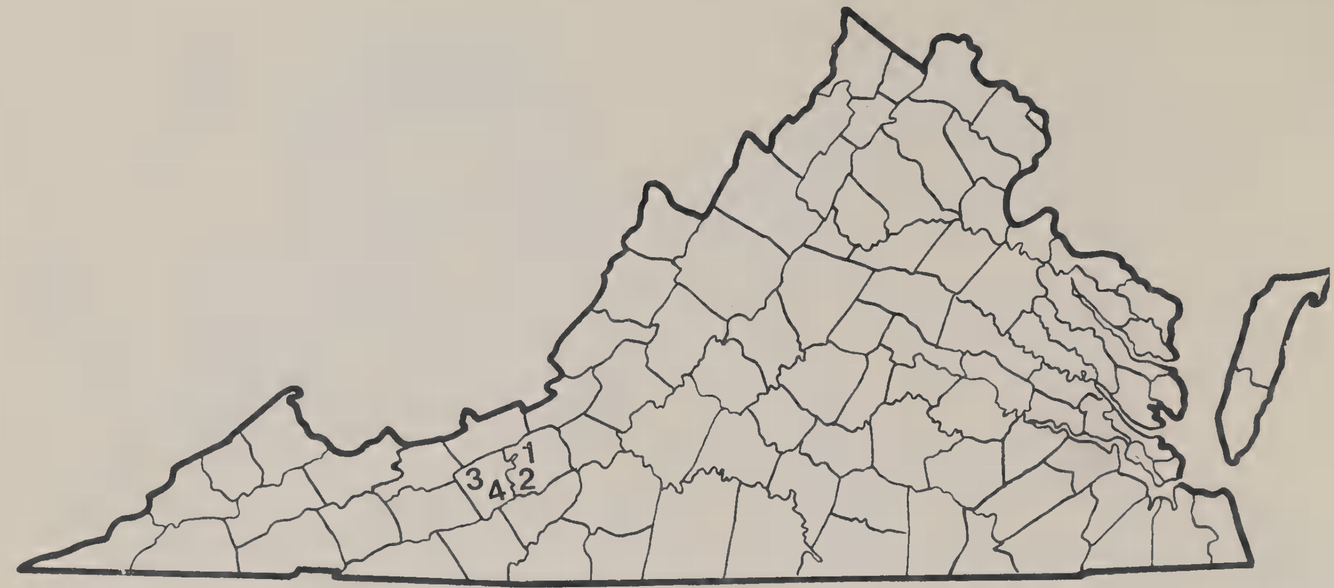

Figure 3.--Releases of C. *horridus* on musk thistle in Virginia. 1 & 2, Montgomery County. 3 & 4, Pulaski County.

TABLE 3.--*Ceuthorrhynchidius horridus* on musk thistle in Virginia

Date	Phase of work
1970	Initial introduction under quarantine
1972-74	Host specificity testing
1974/75	Field releases at 4 sites
1977	Recovery and initial establishment at 4 sites

TABLE 4.--Initial establishment of C. *horridus* on musk thistle in Virginia

Location (County)	Date of release	# of weevils	Recovery of larvae 1976	Recovery of larvae 1977
Montgomery 1	Oct 1974	2,000 larvae & 30 adults	*	*
	Apr 1975	50 adults		
Montgomery 2	Nov 1975	100 adults	*	*
Pulaski 1	Nov 1975	100 adults	-	*
Pulaski 2	Nov 1975	100 adults	-	*

*: Recovery of larvae in rosettes.

-: No recovery.

TABLE 5.--*Ceutorhynchus trimaculatus* and *Cassida rubiginosa* on musk thistle in Virginia

Species	Date	Phase of work
Ceutorhynchus trimaculatus	1975	Initial introduction under quarantine
	1976/77	Host specificity testing & biological studies
Cassida rubiginosa	1973	Initial detection in northern Virginia
	1974/75	Biological studies and field release in Southwest Virginia
	1976	Establishment and recovery in Southwest Virginia

in a dense patch of rosettes and larvae were inoculated into the growth points (punctured by forceps) with a fine camel-hair brush. This was also the first release of this European weevil in North America. The other 3 releases of adults were made in 1975 (Table 4). Detection surveys conducted annually in March and April showed initial establishment in all 4 sites by the spring of 1977 (Kok and Trumble 1977); larvae were recovered in the meristematic tissues of the rosettes. This indicates its ability to survive in Virginia. Its potential for thistle control is being evaluated. Since it attacks young rosettes, it does not directly compete for the same feeding site as *R. conicus*. The two species can thus be used simultaneously in the same area.

Ceutorhynchus trimaculatus, a third curculionid, is another rosette feeder that is currently being screened in our laboratory. The initial shipment was received for quarantine testing in the fall of 1975 and a second in the spring of 1976 (Table 5). Under a cooperative project with the USDA laboratory at Albany, California, host specificity tests and biological studies were initiated in 1976 and it is currently in the final stages of testing. The objective of this project is to determine its safety for field release in the U.S.A. Preliminary adult starvation and oviposition tests conducted by Dunn in Rome indicated that it has potential as a biotic agent. Our tests have been concentrated on development of larvae, oviposition and oogenesis on selected hosts along the same lines as *C. horridus* (Ward *et al.* 1974, Kok 1975a). Results thus far indicate that a few may complete development on *Cynara scolymus* although it shows a distinct preference for the various thistles. It has potential as a biological control agent in Virginia.

Various phases of the work on *Cassida rubiginosa* are shown in Table 5. *C. rubiginosa* was found on musk, plumeless and Canada thistle in Northern Virginia (Kok 1975b; Ward 1976; Pienkowski and Kok 1977). This leaf-feeding insect was accidentally introduced into North America from Europe and was first discovered in Quebec, Canada in 1901 (Fyles 1902). From Quebec, it has

spread across much of eastern Canada and northeastern U.S.A. Although Zwölfer and Eichhorn (1966) showed that *C. rubiginosa* would attack artichoke, *Cynara scolymus*, it has not been reported as being a pest of any economic plant grown in northeastern North America. Ward (1976) conducted biological studies of this insect and found it to be attacked by 5 species of parasites, of which *Tetrastichus rhosaces* and *Eucelatoriopsis dimmocki* were in significant numbers. He isolated several groups of *C. rubiginosa* from its major obligate parasites and released them in southwest Virginia. The objective was to determine whether this beetle will become a more significant thistle-regulating agent in the absence of its parasites. Future evaluations after the beetle has become well established will be necessary to provide the answer. As a foliage feeder, it should place additional stress on thistles and be complementary to the influence of the head weevil and rosette weevil.

All the above species of beetles possess many desirable qualities essential for biocontrol agents: effectiveness, host specificity, high reproductive capacity and good host-seeking ability. Since each insect attacks a different portion of its host, it will be of considerable advantage to have more than one biological control agent acting against the thistles. The combined use of several biological agents would exert pressure on the thistles over a greater proportion of their life cycle and should increase the efficiency of biological control by shortening the interval between establishment and noticeable impact on the thistles. Thus, the chances for efficient and economical control of thistles should be better with a complex of biological agents than with one species. Evaluation of the combined impact of *C. horridus* and *R. conicus* on both musk and plumeless thistle was initiated recently under a cooperative agreement with the USDA-ARS at Beltsville, MD. Four sites were selected for the establishment of both weevils; the thistle densities were determined prior to the beetle releases and will be monitored in relation to the population dynamics of the weevils. Although we have made significant progress towards finding an environmentally acceptable solution for the control of musk thistle, greater efforts will have to be made if the overall thistle problem is to be solved. The presence of several thistle species makes this task more difficult. Successful use of biological control agents will reduce the environmental hazards caused by continued use of herbicides. Finding a permanent solution to these noxious weeds in the pastures of North America will mean better pastures for livestock, benefiting not only the livestock industries, but ultimately the consumers.

ACKNOWLEDGEMENTS

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(c) Virginia State Funds and

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NOTES

[1]Resembles *Carduus thoermeri*.

[2]Previously published as *Ceuthorhynchidius horridus* (Coleoptera: Curculionidæ).

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INTERACTIONS OF *RHINOCYLLUS CONICUS* AND THISTLES IN THE GALLATIN VALLEY

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The Gallatin Valley, located in southwestern Montana, covers an area of approximately 2,300 km^2, and ranges in elevation from 1,268 m at the western side to 1,480 m at Bozeman. Average precipitation in the Gallatin Valley for 1974 through 1976 was 43.5 cm/year, and the mean temperature was 5.9° C, with a maximum of 37° C and a minimum of -35.6° C.

Of the 9 species of *Cirsium* and *Carduus* thistles in the Gallatin Valley, *Rhinocyllus conicus* (Froelich) has utilized 4. Only 1 of these is native, wavyleaf thistle [*Cirsium undulatum* (Nutt.)][1]. Canada thistle [*C. arvense* (L.) Scop.] was first collected in Montana in 1900, and near Bozeman in 1902. Bull thistle [*C. vulgare* (Savi.)] was also collected in Montana in 1900, but was not recorded from the Gallatin Valley until 1955. First records of musk thistle (*Carduus nutans* L.) are from Lolo, Montana in 1922, and from the Gallatin Valley in 1952[2].

Both wavyleaf and bull thistles occur on prairies, grasslands, and pastures and along roadsides, but neither is found in large numbers in the Gallatin Valley and they therefore are of little importance. Both Canada and musk thistles, however, are very abundant throughout the valley, occurring almost everywhere except on newly cultivated or densely forested land.

METHODS AND MATERIALS

R. conicus for release was obtained from France and Italy through the Beneficial Insect Introduction Laboratory at Albany, California. All original releases in Montana were made by or under the direction of the late Dr. Jesse Hodgson. A total of 2,940 adult weevils were released at 6 different intervals at 5 sites between 1969 and 1973. Following each release, observations were made only to confirm establishment; a complete survey of the Gallatin Valley was not made until July of 1974. At that time, musk thistle plants at 224 sites were examined for the presence of *R. conicus*, and other thistle species were examined when encountered. Seed heads of musk thistle from many of these sites were collected for laboratory examination[3, 4].

In 1975, 14 permanent sites were established to study the effects of *R. conicus* on musk thistle. These were sampled regularly at 10-14 day intervals during the 1975 through 1977 growing seasons. At least 20 seed heads and some stem material were collected during each stop and taken to the laboratory for cell and life stage counts, measurements, and rearing of weevils and parasites. Counts of seedling thistles were also made at these sites each fall

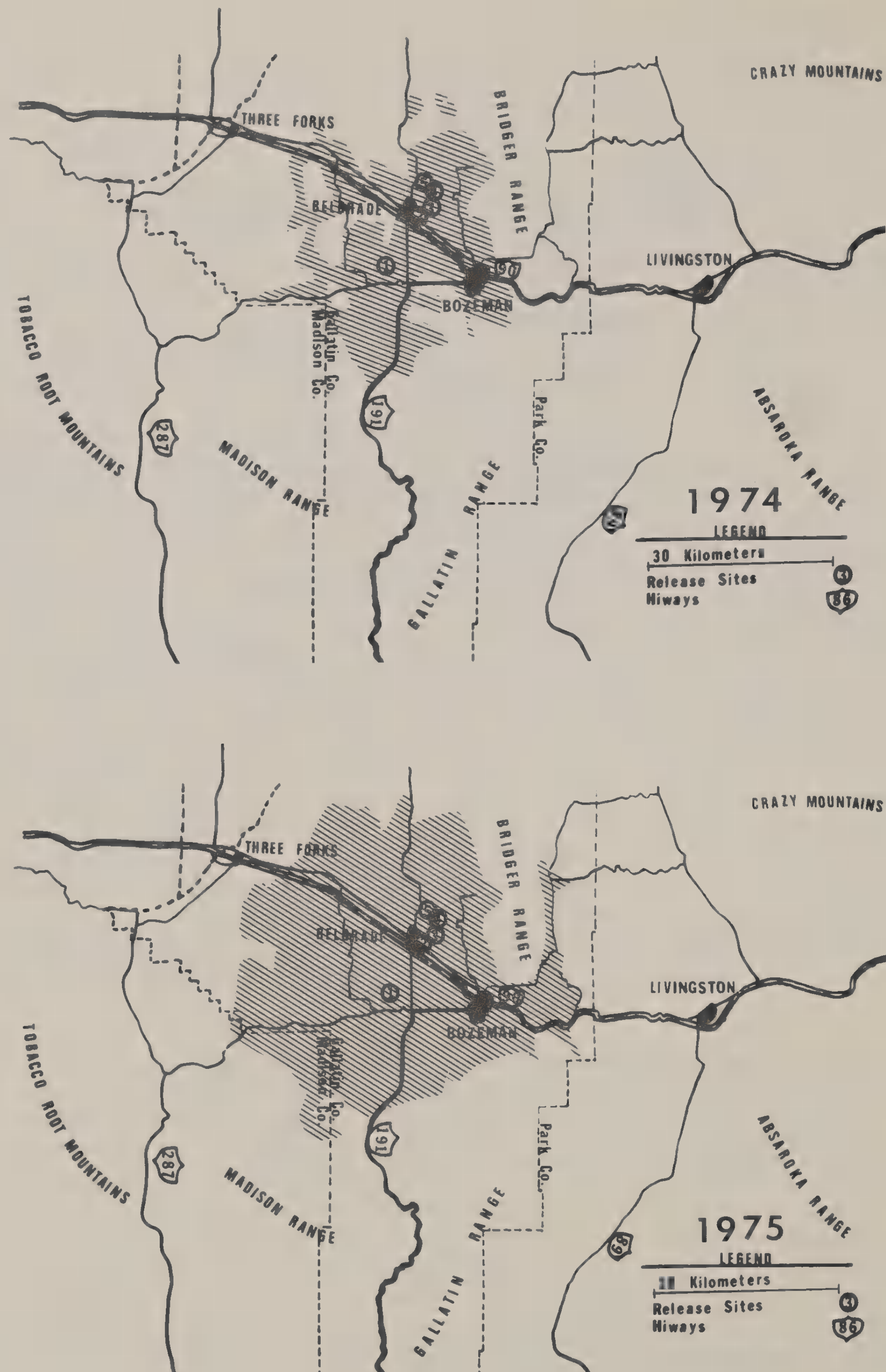
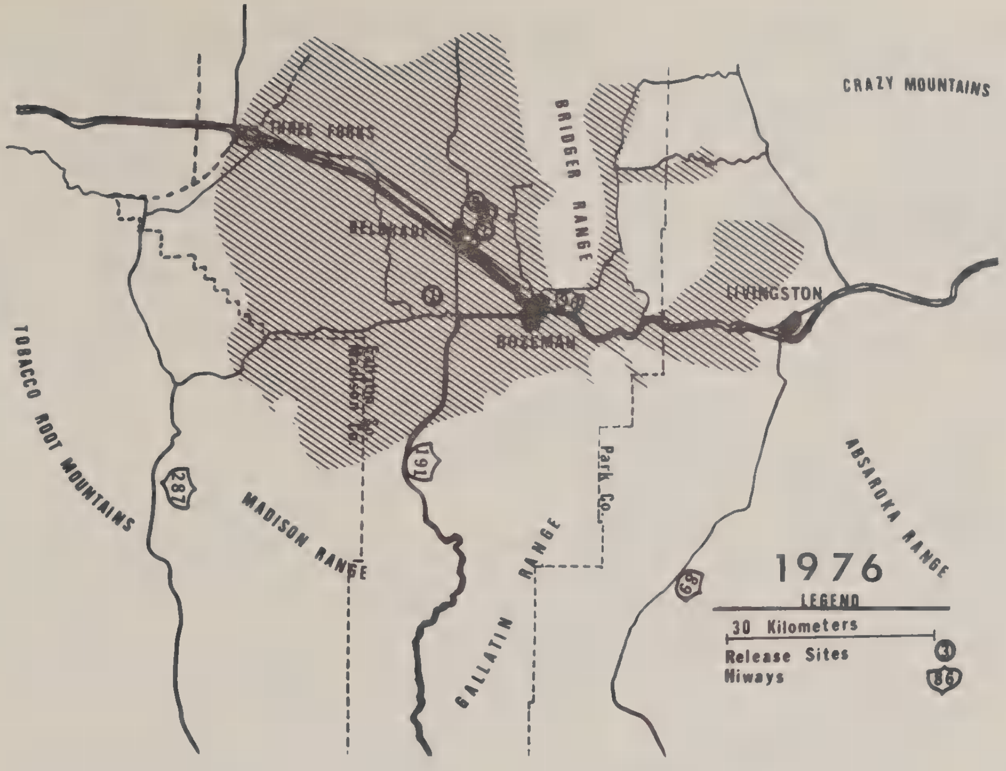
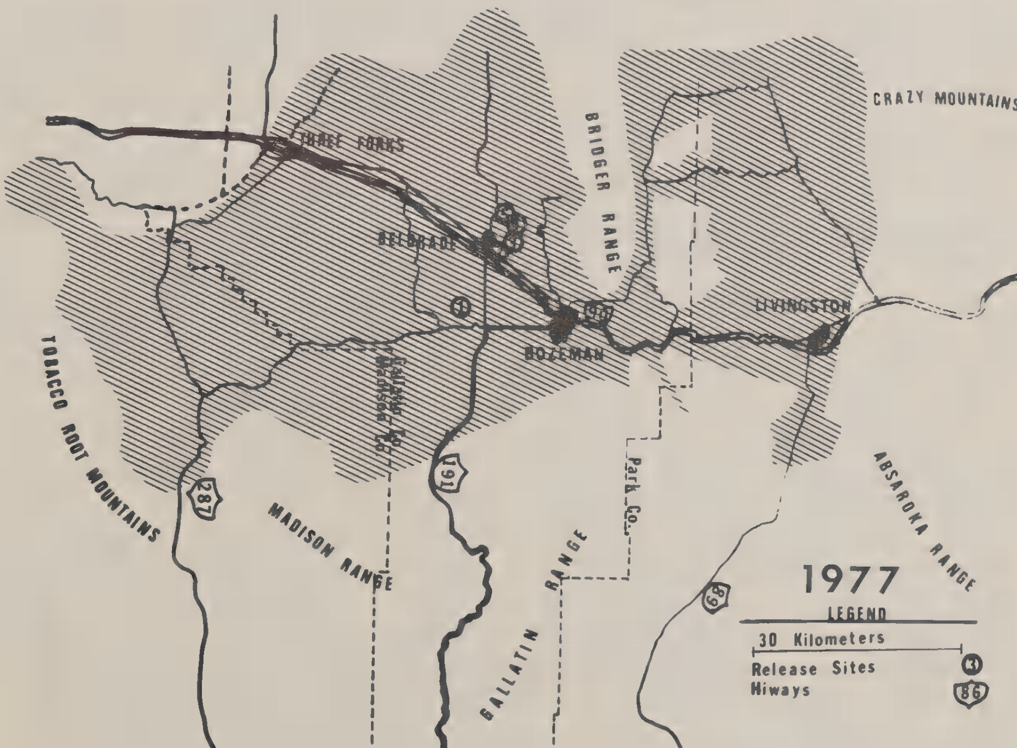


Figure 1.--Expansion of *Rhinocyllus*





conicus population by years, 1974-77.

and spring. Additional sites were inspected intermittently, and annual surveys were conducted to establish migration boundaries of the weevil.

During 1975, 10 sites containing Canada thistle were established to study effects of *R. conicus* on that weed. Ten sites each were also established in 1976 for wavyleaf and bull thistles after larvae had been discovered in those species the previous year. Collections were similar to those of musk thistle, except that no seedling counts of these 3 thistle species were made and 100 seed heads per site was the minimum sample size for Canada thistle. Although additional thistle species were examined, none bore eggs or larvae of *R. conicus* so no test plots for them were established.

RESULTS AND DISCUSSION

The release sites were along a 16 km line from southwest to northeast. Apparently all 6 releases were successful, even though sites 1 and 2 were sprayed with 2,4-D [(2,4-dichlorophenoxy)acetic acid] in 1970 and 1971, respectively. Prior to 1974, annual monitoring of release sites did not indicate a great increase of the insect population, since only a few infested musk thistle seed heads were found each year. However, a survey in 1974 revealed that *R. conicus* was present in musk thistle seed heads in an area roughly 1280 km^2. By the summer of 1977, the boundaries of *R. conicus* had expanded out of the Gallatin Valley and covered an area of about 7,296 km^2 (Fig.1).

Density and mortality figures obtained from the 14 permanent musk thistle sites during 1974 through 1977 are given in Table 1. The mean density of eggs per primary flower increased to 133.0 in 1976, and essentially maintained this average in 1977. The average number of older larvae per flower increased each year without causing an increase in percent mortality. In 1976, mating beetles were observed between May 21 and June 28, which was an increase of 28 days over the 10-day mating period of 1975. This was associated with an increase in the percent of later flowers being parasitized. A decrease of 12 days in the oviposition period during 1977 was accompanied by a decrease in the percent of later flowers parasitized.

Over the 4-year period, progressively higher numbers of adult weevils in the spring were associated with saturation of musk thistle flowers with weevil eggs, higher numbers of larvae per seed head, stems being utilized as oviposition sites, longer oviposition period, oviposition on later musk thistle flowers, higher numbers of eggs deposited on other thistle species, and migration of the adult weevil into unoccupied areas. High numbers of larvae in musk thistle stems often caused death of flowers and subsequent death of many developing larvae in those seed heads.

Causes of mortality of *R. conicus* in and on musk thistle were: (1) egg deposition in excess of available receptacle space, (2) premature drying of the receptacle during larval development, and to a lesser extent, (3) predation and parasitism. Approximately an 80% loss of potential weevil population during 1976 and 1977 was due to over-crowding on the plant surface, whereas among weevils that became established inside of the plant, mortality was only 2-3%. Accurate mortality of larvae within the stems could not be ascertained since cells were not produced in the stem as they were in the seed head, and the higher number of predators and scavengers in the stems destroyed the individuality of many of the remains. Mortality from both parasites and predators was estimated below 1% in the seed heads and stems (Rees, unpublished data).

TABLE 1.--Density and mortality of *Rhinocyllus conicus* on and in musk thistle plants in the Gallatin Valley during 1974-77

	1974	1975	1976	1977
No. of plants examined	12,440	19,580	19,737	18,774
% plants with eggs on flowers	63	88	98	99
% primary seed heads parasitized	63	88	98	99
% later seed heads parasitized	<1	<1	62	42
Density of eggs per primary flower:				
Mean	-	27	133	134
Standard deviation	-	8	71	91
Highest number observed	32	51	316	528
% of plants with eggs on stem	0	<1	37	39
Density of eggs on stem:				
Mean	-	-	13	12
Highest number observed	0	1	41	63
Density of 3rd-instar larvae in primary seed heads:				
Mean	16	18	21	28
Standard deviation	7	9	11	12
Highest number observed	30	44	48	70
% mortality of eggs and 1st-instar larvae[1/]	-	23	82	79
% mortality within the seed head[2/]	3	3	2	3
% of parasitized seed heads with 100% larval survival	64	68	73	56

[1/] Calculated by comparing number of eggs deposited on flower bracts with number of larvae surviving to 2nd-instar.

[2/] Dead larvae, pupae and adult weevils within the plant after normal emergence time divided by the number of cells within the plant.

Musk thistle density has been on a steady decline since the 1974 survey. Mature plants at the 5 release sites averaged 37.4/m^2 in 1974, while fall seedling counts at the 14 permanent sites for 1975, 1976, and 1977 were 20.2, 11.4, and 9.9/m^2 respectively. The large decline in seedlings in 1976 is believed to have been caused by 2 factors which occurred in 1975. *R. conicus* oviposited on 88% of the primary flowers with a resultant 17.8 larvae per seed head, which was enough larvae to destroy the majority of seeds from this source(3). Although later flowers were missed by *R. conicus*, a population explosion of the native pyrilid moth, *Homeosome electellum* (Hlst.), destroyed 89.53% of these seeds. Fig. 2 shows 2 thistle-infested sites photographed in July of 1975 and 1977.

Weevil density during 1974 was insufficient to cause overcrowding in musk thistle, the preferred host. Therefore, only a few eggs were discovered on Canada thistle, and no eggs were found on any other thistle species. However,

Figure 2.--Reduction of musk thistle numbers at two test sites between 1975 and 1977. "Four Corners" site: a, 1975; b, 1977. "Barnes" site: c, 1975; d, 1977.

TABLE 2.--Density of *Rhinocyllus conicus* eggs on species of *Cirsium*, 1974-77

Species of thistle	1974	1975	1976	1977
Canada thistle:				
Number of plants examined	389	3,704	9,097	6,000
% plants with eggs	1	9	47	48
Wavyleaf thistle:				
Number of plants examined	118	216	517	419
% plants with eggs	0	1	41	22
Bull thistle:				
Number of plants examined	96	194	384	244
% plants with eggs	0	1	60	64

TABLE 3.--Percent larval mortality of *Rhinocyllus conicus* in 4 thistle species 1976

Thistle species	% larval mortality
Musk thistle	2.6
Canada thistle	31
Wavyleaf thistle	54
Bull thistle	62

by 1975, 10% or more of the Canada thistle plants at 3 sites had at least 43% of the examined flowers infested. No infestation of the other thistle species was recorded, although eggs were found on several plants of bull and wavyleaf thistles. By 1976 and 1977, the tremendous number of adult weevils present during the spring was associated with oviposition on Canada, wavyleaf and bull thistles (Table 2).

Up to 48 larvae of *R. conicus* can develop within a musk thistle seed head, several can develop in a wavyleaf thistle seed head, and only 1 weevil can survive in either bull or Canada thistle seed heads. Larval mortality was much higher in the other thistle species than in musk thistle (Table 3). Main cause of mortality in Canada thistle was from larvae feeding through the flower to the outside, which either allowed entrance of predators, or killed the flowers and consequently the larvae. Parasitism was minor although some-

what higher than in musk thistle. Mortality in wavyleaf and bull thistles appears to have been the result of these plants being marginal hosts, since most larvae died prior to the 3rd instar from no obvious cause.

SUMMARY

Of the 9 species of *Cirsium* and *Carduus* in the Gallatin Valley, *R. conicus* prefers, and is best able to survive in musk thistle. As *R. conicus* migrates into new areas, musk thistle is sought out first, and Canada thistle is not utilized until musk thistle plants have been saturated with eggs. Bull and wavyleaf thistles are last to be used as oviposition sites.

Both Canada and wavyleaf thistles primarily reproduce vegetatively, and bull thistle is not a preferred host. Therefore, *R. conicus* will have little effect on their populations. Present conditions in the Gallatin Valley suggest that: (1) most of the seeds from the primary flowers of musk thistle, and a majority of the seeds from the later flowers will be destroyed each year by *R. conicus*, with possible sporadic assistance from *Homeosoma electellum*, (2) a reservoir of musk thistle plants produced from seeds of late blooming flowers which were missed by ovipositing *R. conicus* should be large enough, along with the reservoir in Canada thistle, to maintain a high population of *R. conicus* year after year, and (3) as a result of these factors, musk thistle populations should be maintained at a fairly continuous tolerable level.

ACKNOWLEDGMENTS

I recognize, with deepest gratitude, the contributions to this project by the late Dr. Jesse Hodgson. I also express my gratitude to L. O. Baker, Assist. Prof. Agronomy, Montana State University, who assisted with several of the initial releases; Dr. John H. Rumley, Prof. of Botany and Herbarium Head, Montana State University, for assistance with thistle identification and records; Trudie Doornbos for assistance with the surveys; Delbert E. Barnes, Hughes Spain, S. A. Taylor, Robert Ward, Montana State University, and the other numerous ranchers and farmers throughout the Gallatin Valley who set apart a portion of their land for this study, and who allowed me to monitor thistle and weevil numbers on their lands.

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INITIAL ANALYSES OF RHINOCYLLUS CONICUS (FROELICH) (COL.: CURCULIONIDAE) AS AN INTRODUCED NATURAL ENEMY OF MILK THISTLE (SILYBUM MARIANUM (L.) GAERTNER) AND ITALIAN THISTLE (CARDUUS PYCNOCEPHALUS L.) IN SOUTHERN CALIFORNIA

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Rhinocyllus conicus has been successfully colonized in southern California for the biological control of milk thistle and Italian thistle (Hawkes et al. 1972, Goeden and Ricker 1974, 1977, 1978). Initial colonizations used overwintered weevils personally collected from these thistles in west-central and southeastern Italy, respectively (Goeden and Ricker 1977, 1978). Thriving colonies of R. conicus derived from these separate sources were established at separate locations on the same thistles from which they were collected in Italy. Transfers of overwintered weevils from these established field colonies to new but separate locations within the largely sympatric ranges of milk and Italian thistles in southern California (Goeden 1971, 1974a; Dunn 1976) began in 1973 and 1975, respectively (Goeden and Ricker 1977, 1978).

Field data accumulated since the introduction of R. conicus indicate differences in its performance as a biological control agent on milk vs. Italian thistles. Results obtained with R. conicus in southern California also differ in some respects from results recently reported from Virginia (Kok and Surles 1975) and Montana (Hodgson and Rees 1976, Rees 1977). Our initial findings also suggest that one assumption of Zwölfer (1967) used to justify and gain approval for the introductions of R. conicus into Canada (Harris and Zwölfer 1971) and Virginia (Surles et al. 1974) for the biological control of musk thistle (Carduus nutans L.) and welted thistle (C. acanthoides L.), and another assumption relating to its attack strategy (Zwölfer 1973), need to be modified.

HOST RACES OF R. CONICUS

Zwölfer (1967), on the basis of admittedly incomplete data, suggested that, unlike related Larinus spp. (Zwölfer et al. 1971), R. conicus showed no intraspecific differentiation in its host pattern, i.e., did not exhibit distinct host races. Evidence to the contrary now exists.

The first indication of such intraspecific differences involved the unsuccessful attempt to establish R. conicus on milk thistle in northern California in 1969 with stock presumably derived from musk thistle in Europe (Hawkes et al. 1972). Most of the weevils introduced into Canada and the United States before 1971 originated from collections on musk thistle near Mulhouse in the Rhine Valley of France (Zwölfer 1973). This apparently included the weevils first introduced into northern California in 1969, into

Canada in 1968 (Harris and Zwölfer 1971), into Virginia in 1969 (Surles et al. 1974), and into Montana in 1969 (Hodgson and Rees 1976). When weevils collected from milk thistle in Italy subsequently were colonized on milk thistle in northern and southern California beginning in 1971, R. conicus readily was established (Hawkes et al. 1972, Goeden and Ricker 1974, 1977). This suggests that the weevils from musk thistle differ from the weevils obtained from milk thistle in Europe relative to their compatability for the latter, closely related weed, perhaps sufficiently so as to warrant their designation as distinct races.

During annual surveys of R. conicus abundance at its original colonization site on Italian thistle in See Canyon, Calif., eggs on both Italian and milk thistle capitula were recorded during the peak flowering period of the former species (Goeden and Ricker 1978). Milk thistles were common though much less numerous than Italian thistles at this location. Immediately after Italian thistle was surveyed, the flowerheads of all milk thistles in the release area were carefully examined for R. conicus eggs. The number of flowerheads of milk thistle examined each year ranged from 41 to 163. No R. conicus eggs were found on these milk thistle capitula through 1977, while the incidence and frequency of eggs on Italian thistle capitula continued to increase each year (Table 1). Thus, a strongly defined and persistent ovipositional preference was shown by these weevils derived from Italian thistle for this same host under field conditions. This suggests that this colony is comprised of a strain of R. conicus distinct from the strain concurrently and successfully colonized on milk thistle in southern California (Goeden and Ricker 1977).

Milk-thistle-derived R. conicus have shown a similar though less absolute preference for milk thistle over Italian thistle under field conditions in southern California. Table 2 lists egg counts made at Santa Barbara, Calif., at one of the original colonization sites for weevils obtained in Italy from milk thistle in 1971 and 1972 (Goeden and Ricker 1977). Milk thistle was more common than Italian thistle at this location, but the latter was well represented, and both thistles grew intermixed. Our data show that the capitula of milk thistle were consistently and substantially more favored as oviposition sites by this presumed strain of R. conicus during the last 4 years of the 6-year duration of this colony. This site was removed from pasturage and eventually became overgrown with woody perennials and grasses, and sampling was discontinued after repeated collections of overwintered weevils were removed for redistribution to other locations in 1973-77 (Goeden and Ricker 1977).

Another initial colonization site where both milk and Italian thistles occurred was located at Lake Casitas, Calif. (Goeden and Ricker 1977). Here again, weevils originating on milk thistle in Italy and colonized on this weed in 1972 demonstrated a clearly defined ovipositional preference for milk thistle over Italian thistle during the second, third, and fifth years of this colony's existence (Table 3). Italian thistle was not sampled in 1975. A slight increase in the number of eggs on Italian thistle capitula was recorded in 1976, the second year that R. conicus attacked 90% or more of the milk thistle capitula at this site. No such increase occurred at Santa Barbara (Table 2), where the 90% level was reached sooner, presumably because of the smaller area and more isolated nature of the plant deme involved.

The mechanisms governing these differential ovipositional preferences are unknown and warrant further investigation. Controlled weevil crossings might ellucidate the genetic basis of these interspecific differences. Continued long-term monitoring at these and other colonization sites where both milk

TABLE 1.--Frequency distributions for *R. conicus* eggs on Italian thistle capitula recorded during annual, 2 man-hr, random searches at the initial colonization site for this weevil strain at See Canyon, Calif., during 1973-77

Year	Flowerheads with 0-11 eggs													Egg distributions on infested capitula[a]		
	0	1	2	3	4	5	6	7	8	9	10	11	%	$\bar{x}$	s^2	$s^2/\bar{x}$
1973	2613	57	16	2	5	-	-	-	-	-	--	--	2	1.43	.6724	0.47
1974	3114	29	9	3	1	-	-	-	-	-	--	--	1	1.43	.4006	0.28
1975	1540	51	30	22	7	1	-	-	-	-	--	--	4	1.89	.9821	0.52
1976	1754	31	62	19	7	5	-	1	-	-	--	--	6	2.18	1.1622	0.53
1977	52	79	109	114	74	67	36	19	6	2	2	1	91	3.38	6.3326	1.87
														(3.05	4.0184	1.32)[b]

[a] A "coefficient of dispersion" ($s^2/\bar{x}$) of 1.0 indicates a Poisson (random) distribution. Larger coefficients indicate proportionally more clumped distributions; smaller coefficients, more regular distributions (Southwood 1966, Zwölfer 1973).

[b] Statistics in brackets were calculated using uninfested as well as infested capitula; unbracketed statistics, using infested capitula only.

and Italian thistles occur is envisioned, so as to better assess the permanency of these preferences and further determine whether these preferences change at high weevil densities, or ultimately, at the hopefully lower weevil and host new equilibrium densities. Available data are conflicting and too preliminary in this regard, i.e., Tables 1 and 2 vs. Table 3.

Zwölfer's (1967) contention that *R. conicus*, like other Cleoninae, shows a marked affinity for the composite tribe Cynareae, especially for the closely related genera *Carduus*, *Cirsium*, *Onopordum*, and *Silybum*, is not in question. However, as evidence for races of *R. conicus* having distinct host preferences within this predicted "coerced" host range now exists, one criterion used to support the conclusion that the host pattern of *R. conicus* is highly stable, i.e., the absence of host races, no longer appears valid.

EGG DISPERSION BY *R. CONICUS*

At equilibrium densities in its native habitats on musk thistle in Europe, Zwölfer (1973) demonstrated that *R. conicus* coexisted with 3 intrinsi-

TABLE 2.--Incidence of R. conicus eggs on milk thistle vs. Italian thistle capitula recorded during an aggregate 3 man-hr, annual random survey at the Santa Barbara, Calif., colonization site during 1973-76.

Sampling date	Milk thistle				Italian thistle			
	Flowerheads examined		Eggs recorded		Flowerheads examined		Eggs recorded	
	Total no.	No. with eggs (%)	Total no.	$\bar{x}$/ infested head	Total no.	No. with eggs (%)	Total no.	$\bar{x}$/ infested head
May 9, 1973	208	196 (94)	2109	10.7	1052	64 (6)	92	1.4
May 15, 1974	306	276 (90)	2982	10.8	1092	56 (5)	97	1.7
May 29, 1975	326	186 (57)	1441	7.8	956	79 (8)	107	1.4
June 2, 1976	143	143 (100)	2497	17.5	1149	71 (6)	115	1.6

TABLE 3.--Incidence of R. conicus eggs on milk thistle vs. Italian thistle capitula recorded during an aggregate, 3 man-hr, annual random survey at the Lake Casitas, Calif., colonization site during 1973-76.

Year	Milk thistle				Italian thistle			
	Flowerheads examined		Eggs recorded		Flowerheads examined		Eggs recorded	
	Total no.	No. with eggs (%)	Total no.	$\bar{x}$/infested head	Total no.	No. with eggs (%)	Total no.	$\bar{x}$/infested head
1973	397	126 (32)	623	4.9	1119	1 (0.1)	1	1
1974	342	165 (48)	900	5.5	1254	5 (0.3)	5	1
1975	278	261 (94)	2672	10.2	--	- --	-	-
1976	237	227 (96)	3155	13.9	1246	37 (3)	56	1.5

cally superior (1), flowerhead-infesting insect species by virtue of its relatively high fecundity and the regular distribution of its eggs " . . . over practically all available flowerheads." Rhinocyllus thus "spread the risk of intraspecific competition (a) by depositing its eggs on all available capitula of a single host species or (b) by ovipositing on the capitula of co-occurring host species. Has R. conicus behaved in this manner when colonized in California in the virtual absence of such interspecific competition (Goeden 1971, 1974a, 1974b)? Our field data suggest otherwise.

Reference to the preceding section indicates that neither presumed biotype of R. conicus introduced into southern California has, as yet, substantially employed the survival strategy of "spreading the risk" by ovipositing on both milk and Italian thistles when they co-occur at colonization sites (Tables 2 and 3). At See Canyon, at low initial colony densities, only a small percentage of the available Italian thistle flowerheads were attacked, multiple flowerhead attacks were common, and oviposition was thus strongly clumped (Table 1). However, analysis of annual egg distributions on infested capitula by calculation of "coefficients of dispersion" ($s^2/\overline{x}$, Southwood 1966), the method used by Zwölfer (1973), indicates distributions more even than the Poisson (random) type, until the so-called "explosive" phase of colony increase (Frick 1974) was reached after 4 years (Table 1). Egg dispersal by R. conicus was slightly clumped during 1977 (with and without uninfested heads used in computing the dispersion coefficient).

At Santa Barbara and Lake Casitas, egg distributions by the milk thistle strain of R. conicus consistently were strongly clumped, as indicated by the coefficients of dispersion listed in Table 4. Do these differences in oviposition patterns represent additional evidence of musk vs. milk and Italian thistle strains of R. conicus? More likely, they further document the limitations of predictive analysis of natural enemy action (Harris 1973, Wapshere 1973, Zwölfer 1973).

VALUE OF R. CONICUS AS A BIOLOGICAL CONTROL AGENT IN SOUTHERN CALIFORNIA

In addition to the differences noted above, R. conicus shows differential promise as a biological control agent on milk vs. Italian thistles in southern California. Our field data on milk thistle suggest that R. conicus is a less effective natural enemy of this weed in at least one respect--it causes less reduction in seed production, an especially important characteristic relative to the biological control of annual or biennial weeds (Huffaker 1959). On the other hand, larvae have demonstrated a capacity for appreciable direct seed destruction in Italian thistle capitula (Goeden and Ricker 1978). Limited evidence of additional limited indirect reduction of Italian seed production by R. conicus also exists, though not necessarily as a result of larval-induced, increased seed abortion.

This differential effectiveness apparently results from the different sizes of the capitula of these thistles. On the average, the flowerheads of milk thistle are much larger and contain many more achenes than those of Italian thistle. Table 5 records the diameters of the pappi (receptacles) and seed contents of 983 uninfested mature capitula of milk thistle collected in approximately equal numbers from 5 different locations in southern California. These heads averaged 20.8 mm in their pappus diameter, produced an average of 6.2 viable achenes per mm of pappus diameter, for an average of 124 viable achenes.

TABLE 4.--Distributions of R. conicus eggs on flowerheads of milk thistle at 2 original colonization sites where weevils collected from milk thistle in Italy were successfully colonized in southern California.

Location	Year	Mean no. of eggs/ head[a]	s^2	"Coefficient of dispersion"[b] ($s^2/\bar{x}$)
Santa Barbara	1973	10.14	12.6081	15.6789
	1974	9.75	70.5633	7.2409
	1975	4.71	55.9369	11.8784
	1976	17.46	180.7976	10.3540
Lake Casitas	1973	1.56	12.1801	7.8077
	1974	2.63	16.5966	6.3068
	1975	9.58	77.2869	8.0700
	1976	13.31	119.1306	8.9489

[a] Based on total numbers of flowerheads examined, not just on infested heads as in Tables 2 and 3.

[b] All calculated coefficients a strongly clumped egg distribution (>>1.0) (Southwood 1966, Zwölfer 1973).

The greatest basal external diameters of 500 uninfested mature capitula of Italian thistle collected at random at See Canyon averaged only 7.5$\pm$.03 ($\pm$SE) (range: 3.2-8.9) mm. These capitula contained an average of 11.8$\pm$.19 (range: 0-21) fully developed, presumably viable achenes and 3.8$\pm$.13 (range: 0-18) aborted achenes. Comparable data for 490 capitula collected at a weevil-free site at Cayucos, Calif., were: mean basal diameter, 7.5$\pm$.02 (range: 5.9-9.0) mm; mean number of viable achenes, 16.9$\pm$.16 (range: 0-26); and mean number of aborted achenes, 1.6$\pm$.11 (range: 0-17). Thus, milk thistle capitula averaged at least 3 times the width (more likely, 4 times; compare external and pappus diameters of milk thistle capitula in Table 6) and contained an average of ca. 10 times as many viable achenes as Italian thistle capitula.

The receptacles of milk thistle capitula, accordingly, are much larger than those of Italian thistle. The larvae of R. conicus feed and develop within the capitula of various species of composite thistles (Mellini 1951,

TABLE 5.--Numbers of fully formed, presumably viable achenes and underdeveloped, aborted achenes in 983 mature capitula of milk thistle collected at random in approximately equal numbers from each of 5 locations in southern California.

Capitulum diameter (nearest mm)	No. capitula examined	Mean no. viable achenes (± SD)	Range	Mean no. aborted achenes (± SD)	Range
12	6	55.8±25.77	37-69	21.8±12.98	2-40
13	15	55.7±32.24	7-131	26.4±34.55	1-113
14	20	62.7±17.81	15-96	22.3±19.21	3-65
15	51	62.5±27.84	6-123	25.1±31.48	0-139
16	62	70.4±28.09	14-124	28.7±34.53	0-132
17	73	81.6±35.78	0-145	27.5±35.59	0-152
18	71	97.7±33.07	5-155	18.0±28.40	0-134
19	86	117.9±36.99	21-190	13.4±21.05	0-145
20	108	121.8±35.39	6-219	12.9±15.83	0-94
21	106	131.3±36.45	38-211	10.0±18.49	0-148
22	90	142.5±41.44	4-226	6.6±8.54	0-43
23	72	139.7±40.71	0-215	11.5±24.31	0-159
24	42	156.2±50.16	89-259	8.8±8.56	0-22
25	39	140.9±38.20	66-232	6.9±8.98	0-13
26	49	147.7±47.69	0-294	14.5±37.65	0-207
27	28	161.1±40.78	68-307	7.9±8.21	0-36
28	29	157.8±41.31	19-219	13.6±34.52	0-184
29	14	163.1±38.17	74-238	2.6±3.54	0-12
30	12	150.9±26.91	97-188	13.3±15.90	0-45
31	7	184.9±34.76	137-240	4.0±5.41	0-15
32	4	196.5±29.35	156-208	3.3±0.95	2-4

TABLE 6.--Results of artificially induced, high infestations of 23 milk thistle capitula by *R. conicus* in field cagings at San Marcos, Calif., May 14-June 12, 1975.

	External (mm)	Internal (pappus) (mm)	Total no./flowerhead			Total no. achenes/flowerhead			
				Late-stage		Fully developed		Aborted	
			Eggs	Larvae & pupae	Adults	Scarred	Unscarred	Scarred	Unscarred
$\bar{x}$ =	35	24	81.9	4.4	23.8	79.7	33.7	7.0	57.8
$\pm$SD	4.2	2.4	23.15	5.89	10.53	39.76	19.51	7.10	22.43
range =	25-42	18-28	51-134	0-28	0-44	7-177	5-98	0-29	13-102

Zwölfer 1965, 1967). In hosts with large flowerheads, e.g., milk thistle, larval feeding is mainly confined to the receptacle, but in small-headed thistles, e.g., Italian thistle, the small size of the receptacle promotes larval feeding on the developing achenes as well. Zwölfer (1967) remarked that when larval feeding occurs " . . . within the big heads of Silybum or Onopordum the destruction of achenes appears to be less marked." A measure of this "less marked" achene destruction is provided by the following field data.

Three hundred postblossom mature Italian thistle capitula naturally infested with R. conicus last-instar larvae and pupae from See Canyon were individually caged in the insectary until weevil emergence ended (Goeden and Ricker 1978). Upon dissection, these capitula were found to contain an average of 7.3 fully developed achenes, of which averages of 6.3 (ca. 86%) were destroyed by larval feeding, 0.3 (ca. 4%) had a visibly scarred seed coat, but otherwise appeared intact and viable, and 0.7 (ca. 10%) were unattacked. The scarred achenes probably would not have survived to germinate in nature; therefore, an average of 1.8 (range: 1-6) weevils reared from these capitula destroyed ca. 90% of the fully formed achenes therein (Goeden and Ricker 1978).

No comparable seed destruction was noted in the hundreds of flowerheads of milk thistle annually sampled each year at the initial colonization sites (Goeden and Ricker 1977). During 1975, we conducted field trials to evaluate the potential of R. conicus for seed destruction in milk thistle capitula at artificially induced high rates of weevil infestation.

During May 14-June 12, 1975, 30 young preblossom terminal flowerheads of milk thistle at San Marcos, Calif., were individually caged in situ for 1 week with 4 pairs (1 ♂, 1 ♀) each of mature adult R. conicus. Green-dyed organdy sleeve cages were used which were secured at the bottom with masking tape wound firmly around each peduncle after the weevils were introduced. The cage color camouflaged these cages which were located in a field near a busy highway intersection. The cages caused negligible elevation of flowerhead temperatures. The weevils were removed after 1 week, but the heads remained caged in situ until July 21, when most of the R. conicus had completed their development. The peduncles were then severed well below the cages and the excised heads were taken to the laboratory for temporary storage under refrigeration and subsequent dissection.

The results of dissections of 23 of these flowerheads that bore 50 or more eggs are summarized in Table 6. This egg density was found on only 2 of a total of 1515 infested flowerheads examined during annual egg surveys at 11 milk thistle colonization sites throughout southern California during 1975. Even at these high rates of R. conicus attack, an average of 34±19.5 (±SD) intact, apparently viable achenes still was produced per capitulum (Table 1). An average of 57% of the average total of 178±47.0 fully developed achenes in these capitula was scarred and presumed to be destroyed. Prospects are slight that R. conicus field populations will attain densities needed to achieve widespread and massive flowerhead infestations approaching the 81.9 eggs/head average density attained in this field experiment. None of the capitulum infestations induced with these cagings resulted in total seed destruction, even at the highest density achieved of 134 eggs and 44 weevils/head (which still contained 45 unscarred, viable achenes).

Evidence of increased achene abortion at high larval densities was obtained from these cagings. If the pappus diameters of each caged flowerhead is used to derive an expected number of viable and aborted achenes using the data in Table 5, these caged capitula should have produced an average of 144 viable and 11 aborted achenes. Instead, they contained an average of 113

viable and 65 aborted achenes (Table 6). An average of 87% of the aborted achenes in the caged flowerheads were scarred by R. conicus larvae. Whether this represented a direct cause-and-effect relationship, or the incidental scarring of indirectly aborted seed, is unknown.

Thus, the prospects for biological control of milk thistle by use of R. conicus alone in southern California appear dim, though not hopeless. We do not know what seed production is needed for milk thistle stand maintenance, and R. conicus may yet prove its worth by destroying a small though critical portion of the annual seed crop. Seed viability and longevity and seed mortality from external causes other than R. conicus, e.g., ground-squirrel seed-feeding (Goeden and Ricker 1977), also have not been evaluated in this regard. At present, however, it appears that additional natural enemies will have to be found and introduced (Goeden 1974a), if more than a partial degree of biological control of milk thistle is to be achieved.

RHINOCYLLUS CONICUS IN SOUTHERN CALIFORNIA VS. OTHER STATES

One important difference to be noted in the performance of R. conicus as a biological control agent in southern California relates to the complete control of musk thistle it has afforded in at least 1 location in Virginia (Kok and Surles 1975). Prospects that such a high degree of biological control similarly will occur at any of our colonization sites in 1978 or 1979, 6 years after our initial release of R. conicus (as reported for musk thistle by Kok and Surles (1975)), are not encouraging. No signs of the reduced plant heights, capitula production, or plant densities that heralded the 98+% control reported by Kok and Surles (1975) have yet been noted with milk thistle or Italian thistle.

Other differences noted include the report that 2 braconid and 1 ichneumonid parasites of R. conicus first were detected in Virginia in 1971, only 3 years after its initial introduction (Surles 1974). Rees (1977), on the other hand, reported a low incidence (0.2%) "parasitism" of R. conicus by an undesignated agent(s) in Montana in 1975, 6 years after colonizations began. A low incidence of larval parasitism by an unidentified species of Pteromalidae was first observed in Italian thistle at See Canyon in 1977, 4 years after the release of R. conicus (Goeden and Ricker 1978). A single specimen of an undetermined species of Torymidae was recovered from a large sample of weevil-infested milk thistle capitula from Lake Casitas, 5 years after the colonization of R. conicus at this site. This represented our first and only record of parasitism of the milk thistle strain of R. conicus in southern California (Goeden and Ricker 1977).

Aborted musk thistle capitula that fail to fully develop or form viable, mature achenes have resulted from R. conicus attack in Virginia (Kok and Surles 1975) and Montana (Rees 1977). Larval cells in musk thistle "stems" (peduncles?) were described as "numerous" during 1976 by Rees (1977). No such capitulum abortion nor peduncle attack by larvae has been noted to occur on milk or Italian thistles in southern California.

The presumed strain of R. conicus originating from and colonized on musk thistle in Virginia and Montana has been reported to regularly though less frequently and less successfully attack nodding thistle in Virginia (Surles and Kok 1976, 1977) as well as Canada thistle (Cirsium arvense (L.) Scopoli), bull thistle (Cirsium vulgarae (Savi) Tenore), and wavyleaf thistle (Cirsium undulatum (Nuttal) Sprengel) in Montana (Rees 1977). The release of milk-thistle-derived R. conicus in the same general location in Montana during 1971,

1972, or 1973 (Hodgson and Rees 1976) obscures the strain to which this alternate host attack can be attributed. The strong ovipositional preferences shown by the presumed separate strains of R. conicus from milk and Italian thistles for their respective hosts in southern California to date was described and discussed above.

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(1) In competition with individuals of different species within an infested capitulum (Zwölfer 1973).

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